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Odonates (dragonflies and damselflies) as biological indicators at grazed prairie wetlands

by

Christine Leanne Rice

A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfillment of the

requirements for the degree of Master of Science

in

Wildlife Ecology and Management

Department of Renewable Resources

Edmonton, Alberta

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Dr. F. Schmiegelow

Date approved: Sept 19 03

Abstract

A biological indicator is a surrogate for directly measuring the environment or other harder to survey taxa. Bioindicators provide insights into the ecological ramifications of environmental conditions and complex interactions. Aquatic macro-invertebrates are well established as biological indicators in lotic systems, but little research has been done concerning suitable biological indicator taxa in lentic systems. Odonates (Order Odonata) satisfy most of the selection criteria for suitable bioindicators and have been implicated as being sensitive to environmental degradation in aquatic habitats. The littoral zone is critical habitat for odonates and is also targeted by cattle-grazing, making odonates excellent candidates for indicating the ecological impacts of grazing at prairie wetlands. This study supports the suitability of odonates as biological indicators of the impact of cattle grazing on emergent vegetation structure and the aquatic macro-invertebrate communities at prairie wetlands in south-eastern Alberta, Canada.

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Chapter 1

The Theory and Application of Biological Indicators: A Literature Review

1.1 Biological Indicator Theory

1.1.1 What is a biological indicator?

The fundamental principle behind biological indicator theory is that organisms provide information about their habitats. A biological indicator (or bioindicator) is a taxon selected on the basis of its sensitivity to a particular environmental attribute, and then assessed to make inferences about that attribute. In other words, they are a surrogate for directly measuring abiotic features or other biota. Bioindicators are evaluated through presence/absence, condition, relative abundance, reproductive success, community structure (i.e. composition and diversity), community function (i.e. trophic structure), or any combination thereof (Hellawell 1986, Landres et al. 1988). Communities (i.e. organisms living and interacting with one another in a specific habitat) are generally regarded as the most appropriate indicators for conservation and management since inferences can be made at the ecosystem level, as opposed to being limited to an individual species or population (Kovacs et al. 1992).

Much of the discussion surrounding this theory concerns what exactly it is that biological indicators 'indicate', and whether or not inferences – beyond the condition of that particular taxon – are legitimate. Irrespective of validity, these inferences fall into three categories: 1) the condition of their habitat, 2) population trends of other taxa, or 3) the diversity of other taxa in that habitat, defining three types of biological indicators herein referred to as habitat, population, and biodiversity indicators

respectively (adapted from Caro & O'Doherty 1999). One bioindicator may simultaneously fulfill more than one role.

Flagship and umbrella species are the other two types of surrogate species (Meffe & Carroll 1994, Caro & O'Doherty 1999). Flagships are chosen based solely on their ability to provoke public and political compassion, and serve science by garnering public support for conservation efforts (Caro & O'Doherty 1999). A classic example is the World Wide Fund for Nature's (WWF) adoption of the Giant Panda (*Ailuropoda melanoleuca*) as a symbol of international conservation due its massive public appeal. Problems arise when limitations are ignored and flagships are deemed suitable bioindicators based on popular opinion rather than scientific merit (Simberloff 1998, Andelman & Fagan 2000). This improper application of a flagship species is a misuse of limited conservation resources that serves to discredit single-species management in general. Species that require vast areas of habitat (e.g. Grizzly Bear, *Ursus arctos horribilis*; Caribou, *Rangifer tarandus*; or Green Sea Turtles, *Chelonia mydas*) are often selected as umbrella species and used to delineate protected area boundaries with the supposition that providing habitat for the species that demands the most area will automatically provide for the rest. This approach is criticized for relying more on blind faith than science, since it overlooks both sensitive species and species at risk (Simberloff 1998, Schwartz 1999, Andelman & Fagan 2000).

1.1.2 Why indicator species?

Bioindicators make a broad and intangible concept such as 'biodiversity' or 'ecosystem monitoring' manageable by breaking it down into a specific set of quantifiable indicators (Noss 1990). Inference through biological indicators replaces direct measurement when such measurements are not possible, too expensive/difficult, or too direct (Landres et al. 1988, Caro & O'Doherty 1999). Some historical events are impossible to observe directly, but can be inferred via bioindicators, such as reconstruction of lake pH using diatom communities in lake sediments from known dates (Renburg & Hellburg 1982), or examination of museum bird skins as bioindicators of past mercury concentrations (Thompson et al. 1992). Infinite natural complexity and finite management resources dictate that certain parts must be chosen as surrogates for studying the whole; it is impossible to measure every single abiotic and biotic attribute. Biological indicator theory serves to select surrogates that offer managers the largest universe of extrapolation.

Environmental conditions are often highly variable, making it too difficult or too expensive to get accurate measurements. Some substances are altered very quickly after they enter the environment and may be easily missed by infrequent measurements. It may not be cost effective to monitor as frequently as is needed to obtain an accurate reading, especially for brief (e.g. industrial releases) or unpredictable intermittent events (e.g. sewer overflows, storm run-off). Measuring water quality is a good example since water chemistry is inherently so variable; there are temporal and spatial challenges with data collection; measurements only really provide information on conditions at that moment in time (Spellerburg 1991, Resh et

al. 1996). By monitoring organisms in addition to physical/chemical attributes a longer temporal aspect is introduced since organisms incorporate past, as well as present, conditions (Rosenburg & Resh 1996).

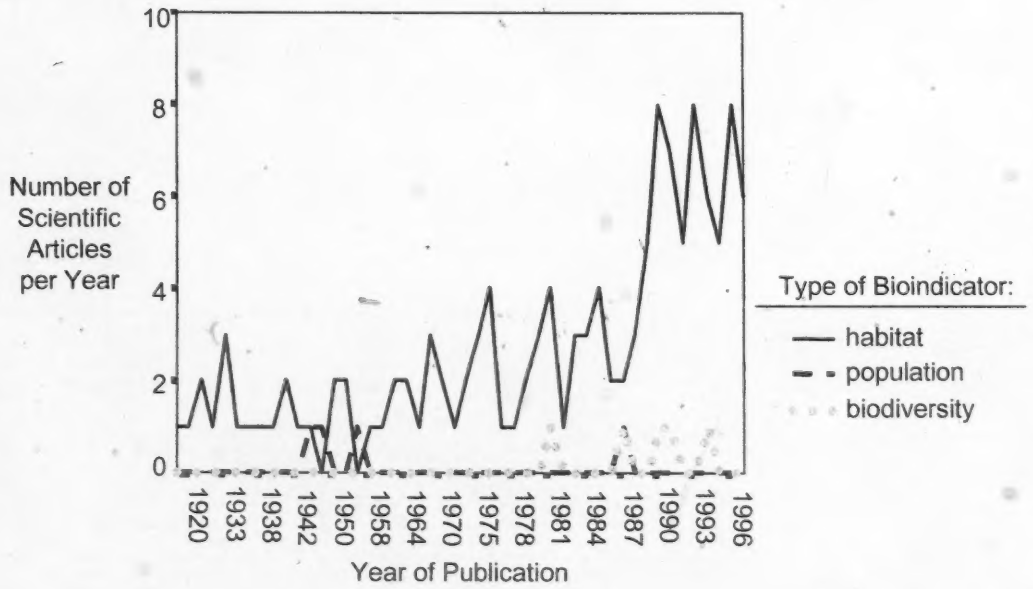
Direct measurements of abiotic (e.g. pollution) or biotic (e.g. introduced species) variables are important but also too direct to provide insight into ecological effects, especially when considering the synergistic effects of multiple factors; we lack complete understanding of synergistic interactions and often of the appropriate choice of substances to measure (Karr 1981, Croonquist & Brooks 1991, Karr 1991, Spellerburg 1991, Kremen 1992, Lambeck 1997, Paoletti 1999, Hilty & Merenlender 2000, US EPA 2002a). For example, mercury concentrations in arctic ice are too low to get an accurate (uncontaminated) reading, yet unhealthy concentrations are found in porpoises, birds, humans and bears due to bioaccumulation (Thompson et al. 1992). Measuring abiotic parameters exclusive of the impact of these conditions on biotic indicators may provide incomplete or inaccurate information on the state of an ecosystem.

1.1.3 History of biological indicator theory

The term "bioindicator species" was coined by Kolkwitz and Marsson in 1908 and 1909 regarding the impact of organic pollution (i.e. sewage) on aquatic organisms (Rosenburg & Resh 1996). Bioindicator literature has since developed to include the concepts of population and biodiversity indicators (originating around 1942 and 1980, respectively), though the bulk of articles remain on the topic of habitat indicators (Figure 1–1; Appendix-A).

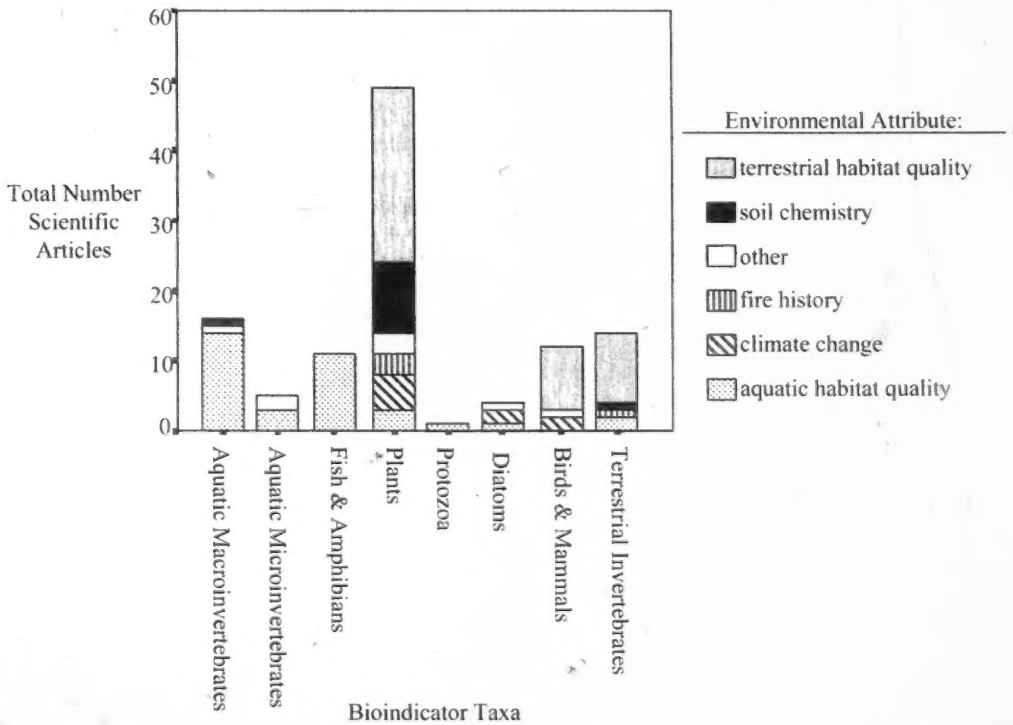
Habitat indicators are definitely the most accepted and studied type of bioindicator in the scientific literature. This concept arose in the field of botany, therefore the bulk of the early literature deals with plants as indicators of soil chemistry or habitat quality (i.e. pollution), but other commonly studied taxa include vertebrates and aquatic invertebrates (Figure 1–2; Appendix-A). Scientific merit, not frequency of use, determines the suitability of a bioindicator and the most recommended taxa for bioindicator use are aquatic macroinvertebrates and algae (e.g.. recommended by 27% and 25% of the reviewed articles; other taxa included protozoa, bacteria, fish, macrophytes, fungi, yeasts, and viruses) (Hellawell 1986).

Figure1–1: Timeline of the types of biological indicators discussed in the scientific literature during the 20th century (earliest publication to 1998).
Results of a JSTOR© database search (Appendix-A for details).



Indices of Biotic Integrity (IBI) and other similar multi-metric concepts have developed since the 1970's as methods to quantitatively assess environmental condition through habitat indicators (Rosenburg & Resh 1996). IBI's are an alternative to conventional statistical analyses that aim to condense and clarify complicated ecosystem interactions into a numerical index based on scores for "high" or "low" environmental quality from various metrics (i.e. habitat indicators; usually 8-12 per site) (US EPA 2002a). Karr (1981) coined the method for quantitative assessments that has since been widely applied and developed as a tool to evaluate riparian health (almost exclusively in lotic systems) by many state and federal agencies in the U.S.A., most prominently the U.S. Environmental Protection Agency (US EPA 2002a).

Figure 1–2: Distribution of scientific articles (earliest – 1998) among different bioindicator taxa. Results of a JSTOR© database search (Appendix-A).



Biological indicators are currently used and promoted by numerous conservation agencies as a means to tackle biological monitoring and assess human impacts, including the World Conservation Union (IUCN), World Conservation Monitoring Centre (UNEP), U.S. Environmental Protection Agency (US EPA), as well as the Nature Conservancy, World Wide Fund for Nature (WWF), Friends of the Earth (FOE), and Greenpeace (IUCN 1989, US EPA 2002a, UNEP 2002).

1.1.4 Critique of biological indicator theory

Biological indicators have high intuitive appeal and have been eagerly embraced by many conservation managers, frequently without regard for selection criteria or empirical evidence (McGeoch & Chown 1998). The most common criticism of indicator species is that scientifically invalid criteria have been used for their selection, whether this is socio-economic pressure to study charismatic macro-fauna (Landres et al. 1988, Niemi et al. 1997, Simberloff 1998, Hilty & Merenlender 2000), or the desire to study a convenient or favourite taxon (Williams & Gaston 1994, Stork & Samways 1995, McGeoch 1998). A lack of standardized techniques for bioindicator-based research has been implicated as a reason for controversy with respect to its validity (Reyers et al. 2000). These criticisms can be addressed by basing indicator taxa selection on research objectives, rather than vice versa, and by following the guidelines and selection criteria offered in the literature (Phillips 1980, Hellawell 1986, Pearson 1994, McGeoch 1998, Caro & O'Doherty 1999, New 1999, US EPA 2002b). Odonates were chosen as bioindicators for this thesis based on published research on their sensitivity to environmental disturbances in lotic systems

(Watson et al. 1982, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998), and the results of preliminary research within the study area in 1999 (Hornung & Rice 2003).

Even when adequately selected, controversy remains over what exactly it is that indicators 'indicate' (i.e. what is the legitimate universe of extrapolation?). Some argue that biological indicators only provide information about that particular taxon and it is conjecture to extrapolate findings to other taxa (Simberloff 1998). Others argue that it is perfectly reasonable to infer how other taxa are faring once a relationship has been established between indicator and indicatee (McGeoch 1998, Caro & O'Doherty 1999). The ramification is that this relationship must be established for each region (i.e. at least each continent – see Pearson & Carroll 1997) and system (e.g. lotic vs. lentic aquatic habitats), and this need for empirical confirmation diminishes the practical appeal of bioindicators as management short-cuts, at least in the short-term.

Each of the three types of biological indicators is critiqued further below.

1.1.5 Habitat indicator species

Habitat indicators provide information about the quality of their habitat through their physical condition or presence/absence, thereby effectively functioning as "biological litmus paper" (Hellawell 1986). There is little controversy concerning their validity since they involve very little inference and have a long history of study in the fields of botany and environmental toxicology (Landres et al. 1998, Paoletti 1999). The scientific community appears to accept the need to assess how changes in

the physical environment are reflected in the biotic community (Karr 1981, Landres et al. 1988, Karr 1991, Paoletti 1999, US EPA 2002a).

1.1.6 Population indicator species

Population indicator species are also selected based on their sensitivity to particular environmental attributes. An important distinction is that their population trends are extrapolated to reflect those of similar species, rather than the condition of their environment (Landres et al. 1988, McGeoch 1998, Caro & O'Doherty 1999). This "guild-indicator approach" is charged with oversimplifying interspecific relationships since species are different in their response to habitat changes and mechanisms of population control (e.g. sensitivity to disease resistance), and the removal or decline of one species may actually benefit a similar species by freeing up limited resources (Landres et al. 1988, Caro & O'Doherty 1999). To ensure the validity of population indicators the relationship between the indicator species and its guild members must be empirically established. Little correlation has been shown to exist between indicator taxa and guild members (e.g. forest bird communities; Mannan et al. 1984, Szaro et al. 1986), except concerning predator – prey relationships (e.g. fewer Cape Gannets (*Morus capensis*) when anchovies were scarce (*Engraulis capensis*), or correlations between abundance of White-backed Woodpeckers (*Dendrocopos leucotos*) and saproxylic beetles (Order Coleoptera); Oatley et al. 1992 and Martikainen et al. 1998, respectively).

1.1.7 Biodiversity indicator species

The most recent application of bioindicator theory developed from the concept of "biological diversity", which was made commonplace by E.O. Wilson in the late 1980's (Wilson 1988) and the Rio Earth Summit in 1992 (UNEP 1992) (Figure 1–2). There is much discussion in the scientific literature as to the validity of biodiversity indicators, with studies that both support (Pearson 1992, Kremen 1994, Pearson 1994, Carroll and Pearson 1998, and Reyers et al. 2000) and oppose this theory (Landres et al. 1988, Kremen 1992, Prendergast 1993, Faith & Walker 1996, and van Jaarsveld et al. 1998). Biodiversity indicators have been used to infer lower taxon (i.e. species) richness by surveying higher taxonomic levels (i.e. family) (supported by Oliver & Beattie 1993, Gaston 2000; disputed by Goldstein 1997, Prendergast and Eversham 1997), however they are more commonly used to identify hubs of biodiversity by inferring overall diversity from that of an indicator taxa (McGeoch 1998, Caro & O'Doherty 1999).

The ability to infer overall biodiversity from a single index is disputed (Landres et al. 1988, Simberloff 1998, Faith & Walker 1996, van Jaarsveld et al. 1998) and has been shown to be invalid in some cases (Goldstein 1997). For example, Oliver & Beattie (1992) found invertebrate diversity inaccurately represents vertebrate diversity, Kremen (1992) found Malagasy butterflies were inappropriate indicators of plant diversity, while Prendergast et al. (1993) found little correlation between the species richness of birds, butterflies, dragonflies, and aquatic plants in Britain. In addition, the presence of threatened or endangered species does not necessarily coincide with areas of high biodiversity, consequently making species at

risk inadequate biodiversity indicators (Bonn et al. 2002), and biodiversity indicators poor indicators of rare or endemic species (Reyers et al. 2000). However, correlations between the diversity of some related taxa do exist though these may be valid only within a particular geographic area, such as a continent or eco-region (Pearson & Cassola 1992, Flather et al. 1997, Pearson & Carroll 1998, Anderson & Vondracek 1999). Research in support of biodiversity indicator theory usually has a very limited realm of extrapolation, such as using one butterfly genus to indicate overall butterfly diversity (Kremen 1994).

Relationships between taxa must be validated rather than assumed in order to accurately identify biodiversity indicator taxon (Prendergast 1993, McGeoch 1998, Sahlen & Ekestubbe 2001), and more than one taxon should be used to infer overall biodiversity and avoid great oversimplification (Wilson 1988, Croonquist & Brooks 1991, Kremen 1992 and 1994, Kremen et al. 1993, Saetersdal & Birks 1993, Launer & Murphy 1994, Lambeck 1997, Caro & O'Doherty 1999). The need for multiple taxa and validation within each region erases the initial intuitive appeal of bioindicators as a quick and easy answer for assessing biodiversity. Indeed, the initial stages of identifying, testing, and selecting habitat, population, or biodiversity indicators will be (temporarily) resource intensive, but this is a necessary first step in order to ensure the validity of the indicator taxon and the accuracy of subsequent inferences.

1.2 Introduction to the study

1.2.1 Cattle grazing and the prairie landscape

Wetland management on the prairies has increased in intensity in recent years due to the enormity of historical wetland loss, and the prevalence of drought. Conservation efforts have focused almost exclusively on water birds, with invertebrates largely ignored except as food sources for waterfowl and shorebirds (e.g. Chura 1961, Ashley et al. 2000). This is changing since the ecological and intrinsic value of invertebrates, (especially large charismatic taxa like odonates and butterflies), is gaining appreciation among the public and, concurrently, wetland managers. For example, odonates are now included on species inventory and species at risk lists internationally (Moore 1997), nationally (NCC 2001), and provincially (Rice 2000, ANHIC 2002).

Cattle (*Bos taurus*) are given free access to prairie wetlands in the study area as a source of drinking water and supplementary forage. Cattle were commonly observed targeting their grazing efforts on wetland vegetation and effectively removing most of the emergent vegetation (e.g. *Typha latifolia* and *Scirpus* spp.). Unrestricted cattle access to wetlands is a well-established management strategy that has arisen out of convenience and economy rather than ecological study. Conversely, it has been demonstrated that keeping cattle out of wetlands and providing on-site watering troughs can improve cattle health and weight gain, and in turn increase rancher's profits (Willms et al. 1994).

1.2.2 Aquatic invertebrates as bioindicators

Invertebrates are generally regarded as better bioindicator candidates than vertebrates due to their diversity, relatively quick response to environmental change, and inability to adapt outside of particular physiological constraints (Pearson & Cassola 1992, Kremen et al. 1993, Weaver 1995, but see Mensing et al. 1998). Sedentary invertebrates can be used to identify the location of a particular environmental stress, long-lived (i.e. years) species facilitate studying the effects of sporadic disturbances, and collecting invertebrate samples is simple, inexpensive (e.g. dip nets), and publicly acceptable (Hellawell 1986, Resh et al. 1996). The disadvantages to studying invertebrates are mainly logistical (i.e. laboratory work is time consuming, laborious, and requires a certain level of expertise; Hellawell 1986), and ecological; the inherently patchy distribution of some invertebrates may pose sampling problems, and invertebrate presence or abundance may be confounded by factors other than environmental disturbance (e.g. seasonal variation, substrate availability, dispersal capabilities) (Hellawell 1986, Resh et al. 1996).

Aquatic macroinvertebrates are the most frequently used taxa for monitoring habitat quality (Hellawell 1986), with a well-developed body of information for lotic systems (Hilsenhoff 1987, Kiffney & Clements 1993, Day & Reynoldson 1996, Rosenburg & Resh 1996, Clausen & Biggs 1997, Brooks et al. 1998, Scrimgeour et al. 1998, Barton 2000, Birge et al. 2000, Giggleman & Bocanegra 2000, Barbosa et al. 2001, Braukmann 2001, Lotufu 2001). Most research involves IBI's or similar multi-metric 'rapid assessments' that compare quantitative indices based on numbers and types of species for disturbed versus reference sites, and monitoring programs

employing these techniques are underway in the USA and United Kingdom (Resh et al. 1995, Reynoldson et al. 1997, Karr & Chu 1997, US EPA 2002a & b). IBI's have mainly been developed for specific habitats (i.e. small streams) and pollutants; more research is needed regarding other aquatic habitats (i.e. wetlands) and non-point source pollutants (Rosenburg & Resh 1996).

Research on macroinvertebrates as indicators in lentic systems has only recently begun (i.e. mid to late 1990's) and consequently is far less prevalent in the literature (Resh et al. 1995, Rosenburg & Resh 1996, US EPA 2002a). Procedures for lentic habitat indicators have mainly been adapted from the work of stream biologists (for wetlands see US EPA 2002a; for lakes see Somers et al. 1998 and O'Conner et al. 2000). The US EPA has arguably become the authority on wetland rapid assessments, and has compiled comprehensive on-line information and guidelines for all types of aquatic ecosystems (e.g. wetlands, estuaries, small streams and rivers, lakes and reservoirs, and coral reefs) (US EPA 2002a & c). Otherwise, few studies specifically investigate wetland-tailored IBI's (van Dam et al. 1998, Burton et al. 1999, Kashian & Burton 2000), or macroinvertebrates as wetland habitat indicators (Hicks & Larson 1997, Zimmer et al. 2000, Spieles & Mitsch 2000, and Cohen et al. 2001).

There is a lack of research on aquatic invertebrates as population or biodiversity indicators in both lotic and lentic systems.

1.2.3 Odonates as bioindicators

Odonates are characterized as excellent habitat indicators of present and past (long-term) environmental conditions in aquatic habitats (Watson et al. 1982, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998). Concerning their scientific merit as appropriate bioindicator taxa, odonates satisfy most published selection criteria, rank among the top 20% for all candidate taxa, and are one of the best when considering aquatic taxa alone (Table 1–1; Brown 1991, Clark & Samways 1996).

Table 1-1: Suitability of Order Odonata as bioindicator taxa.
(H=habitat indicator, P=population indicator, B=biodiversity indicator; *=satisfies criteria, ?=unknown. Adapted from Pearson 1994, McGeoch 1998, and Caro & O'Doherty 1999)

		Bioindicator			
	Selection Criteria	**H**	**P**	**B**	**Order Odonata**
Measurement Attributes	Represents other species		*	*	?
	Taxonomically well-known	*	*	*	*
	Easy/cheap to sample	*	*	*	*
	Accessible breeding site	*	*		*
	Single species	*		*	*
	Assemblage of species		*		*
	Baseline population data available	*			not often
Life-history Attributes	Small body size	*			*
	Short generation time	*	*		variable
	High metabolic rate	*			*
Ecological Attributes	Medium home range size	*			*
	Resident	*	*		almost all
	Particular trophic level	*	*		*
Attributes of Commonness	Abundant	*	*		most species
	Ubiquitous	*	*	*	most species
	Habitat specialist			*	*
Environmental Sensitivity	Sensitive to human disturbance	*	*		*
	Low variability in response	*	*		?
Social Attributes	Intrinsic/economic value recognized	*	*	*	*

Odonates inhabit both terrestrial and aquatic habitats during their life cycle and therefore may better reflect disturbance to the riparian buffer than other strict wetland obligates. Despite their suitability, odonates have been employed as habitat indicators relatively infrequently in lotic systems (Carle 1979, Watson et al. 1982, Ferreras Romero 1984, Carchini & Rota 1985, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998), and even less frequently in lentic systems (Chovanec & Raab 1997, Rith-Najarian 1997a & b).

To provide insight into reasons for their response, (e.g. natural or anthropogenic disturbance, organic or inorganic pollution), bioindicators should be monitored in concert with relevant environmental data (Faith & Walker 1996). Adult odonate species richness has been shown to be correlated with macrophyte richness (Rith-Najarian 1997a & b, Stewart & Samways 1998, Painter 1999, Sahlen & Ekestubbe 2001, Hornung & Rice 2003). Declines in odonate richness have been linked to activities that trample and remove vegetation from the littoral zone, including intensive sport fishing (Muller et al. 2003), and wild buffalo (*Syncerus caffer*) trampling (Stewart & Samways 1998).

1.3 Need for this study

This thesis applies a novel approach to researching cattle grazing at prairie wetlands, since environmental data (i.e. water quality and wetland vegetation) are investigated together with long-lived predaceous aquatic invertebrates (i.e. odonates) to provide insight into how the physical impacts of cattle grazing affect wetland biota. Data collected here will help develop bioindicator theory by addressing the gap in

research concerning lentic habitats, population or biodiversity indicators, and non-point source agricultural pollution.

1.4 Thesis outline

This study examines the suitability of odonate community structure as an accurate bioindicator of cattle grazing at prairie wetlands. Chapter 1 provides a literature review of bioindicator theory including its applications and limitations, focussing on macro-invertebrates and aquatic habitats, to provide context and rationale for this thesis.

The following three data chapters test hypothesis concerning specific bio-indicator selection criteria and odonates. The hypotheses presented here are kept broad in order to convey the main topic of each chapter. Chapter 2 investigates odonates as habitat indicators by exploring relationships between larval odonates and cattle impacts on wetland water quality, addressing the following hypotheses:

Hypothesis #1: Cattle affect the water quality of a wetland by urinating and defecating directing into the water.

***Prediction**: Wetlands with greater exposure to cattle grazing (i.e. more continuous grazing regimes) will have poorer water quality than those with truncated or no cattle grazing (as measured by higher concentrations of fecal coliforms, ammonium, nitrates + nitrites, total Kjiedahl nitrogen, total phosphorus, chlorophyll-a, and total dissolved solids).*

Hypothesis #2: The odonate community at a wetland is sensitive to changes in water quality due to cattle grazing.

Prediction: *If cattle grazing decreases wetland water quality, then larval odonate genus composition will be altered and genus richness, abundance, and diversity will be decreased.*

Chapter 3 addresses the impact of cattle grazing on wetland vegetation, and any subsequent impact on the adult odonate fauna of that wetland, with the following hypotheses:

Hypothesis #3: Cattle grazing directly affects the wetland vegetation community.

Prediction: *The vegetation community at wetlands with greater exposure to cattle grazing (i.e. more continuous grazing regimes) will have lower species richness, abundance (i.e. % cover), vertical structure (i.e. average height/species), and diversity.*

Hypothesis #4: Wetland vegetation (providing emergence substrates, perches, oviposition sites, and cover) directly affects the adult odonate community.

Prediction: *If cattle grazing decreases wetland vegetation richness, abundance (i.e. % cover), vertical structure, and diversity, then adult odonate richness, abundance, diversity, and reproductive effort will also decrease, and species composition will be altered.*

Odonates as biodiversity indicators are further investigated in Chapter 4 by examining their accuracy in predicting aquatic macro-invertebrate and potential prey diversity at wetlands with different grazing regimes. The suitability of odonates as bioindicators of the wetland community is assessed by synthesising results from the previous two chapters. The hypotheses tested are:

Hypothesis #5: Larval odonate community structure is an accurate population and biodiversity indicator of the aquatic macroinvertebrate community at a wetland.

Prediction: *If odonate prey and overall aquatic macroinvertebrate richness, diversity, or abundance is low, then larval odonate richness, diversity, or abundance will also be low.*

Hypothesis #6: Odonates are an accurate biological indicator of cattle grazing impacts on the water quality, vegetation structure and diversity, and aquatic macro-invertebrate community of prairie wetlands.

Prediction: *If cattle grazing decreases wetland water quality, then overall larval odonate composition will be altered and taxa richness, abundance, and diversity will decrease.*

Prediction: *If cattle grazing decreases wetland vegetation richness, abundance (i.e. % cover), vertical structure, and diversity, then overall adult odonate composition will be altered and richness, abundance, and diversity will decrease.*

Prediction: *If cattle grazing negatively impacts the aquatic macro-invertebrate community this impact will be mirrored by the larval odonate community.*

Chapter 5 provides a synopsis of odonates as biological indicators at prairie wetlands, and discusses the implications of this study to biological indicator theory and the practical utilization of odonates for prairie bio-monitoring.

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Chapter 2

Larval odonates as bioindicators of cattle grazing and water quality at prairie wetlands

2.1 Introduction

Prairie potholes were formed during the last glaciation on the Great Plains of south-central Canada and north-central United States, and are characteristically small (<50ha) and collectively numerous. More than 50% of these wetlands have been lost, primarily due to agriculture (Mitsch & Gosselink 1993). Ephemeral natural wetlands undergo a 5-20 year drought cycle (van der Valk & Davis 1978), however many remaining potholes are intensively managed for anthropocentric purposes (i.e. drinking reservoirs for cattle, irrigation basins, waterfowl breeding/hunting grounds), effectively truncating natural draw-down periods and decreasing hydrologic variation.

Prairie marshes are typically basic to alkaline, somewhat saline, and highly productive (Mitsch and Gosselink 1993). Nitrogen and phosphorus are the two most important nutrients in freshwater systems, nitrogen being the most limiting, and excessive amounts of either can cause eutrophication (Mitsch and Gosselink 1993). These nutrients enter wetlands via decomposing organic mater, animal wastes (i.e. human sewage, livestock manure), agricultural or industrial run-off, nitrogen fixation, and erosion or re-suspension of phosphorus in parent geological material. Wetlands are generally regarded as nitrogen and phosphorus sinks, and as such, are frequently constructed to amend nutrient loading from organic pollution (i.e. livestock feed lots, human sewage ponds) (Neely & Baker 1989, Mitsch and Gosselink 1993, Peterson 1998).

Grazing is one of the main sources of disturbance on the prairies, and wetlands are utilized by ranchers for the water and vegetation they provide livestock. Prairie wetlands are inherently variable systems that have evolved with wild fire and bison grazing (van der Valk & Davis 1978, Mitsch and Gosselink 1993), and may in fact flourish with some level of disturbance. This study is well suited for testing the Intermediate Disturbance Hypothesis (IDH) (i.e. moderate disturbance increases diversity, Connell 1978) concerning prairie wetlands and cattle grazing. Cattle remove emergent and submergent vegetation, trample shorelines, and deposit feces and urine in and around the wetland. Water quality can be degraded by the addition of fecal coliforms via manure, or increased sedimentation via shoreline trampling and grazing (Meehan & Platts 1978, Mosley et al. 1999). Grazing impacts can be visually significant (Figure 2–1), however, the ecological consequences on wetland nutrients is less apparent, with conflicting research that both confirms (Schepers et al. 1982, Jansen & Robertson 2001, Scrimgeour & Kendall 2002) and refutes (Buckhouse & Gifford 1976, Bohn & Buckhouse 1985, Clark 1998, Nader et al. 1998) cattle's negative impact on water quality.

Figure 2-1: Differences in appearance between intensively grazed and ungrazed prairie wetlands.

Ambiguity regarding the ecological impact of grazing on wetlands may be explained by differences in grazing practices (i.e. intensity, timing, frequency) and landscapes (i.e. vegetation communities, topography) (Clark 1998, Mosley et al. 1999). Soil-water interactions, diurnal or seasonal fluctuations, and storm events further affect the buffering capacity of a wetland (Mitsch & Gosselink 1993, Harker et al. 1998). Furthermore, water chemistry is so inherently variable that any detectable decrease in water quality due to grazing only really provides insight into what conditions exist at the time of data collection rather than the significance of these conditions to the aquatic flora and fauna (Spellerburg 1991, CAST 1992, Resh et al. 1996).

Bio-indicators are valuable for interpreting the ecological consequences of environmental conditions to biota, especially when considering intermittent disturbances (Rosenburg & Resh 1996). Larval odonates have been recognized as sensitive water quality indicators in lotic (Carle 1979, Watson et al. 1982, Carchini & Rota 1985, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998) and lentic systems (Chovanec & Raab 1997, Rith-Najarian 1997a & b), and are prime candidates for investigating the effects of non-point source agricultural pollution (e.g. cattle grazing) at prairie wetlands.

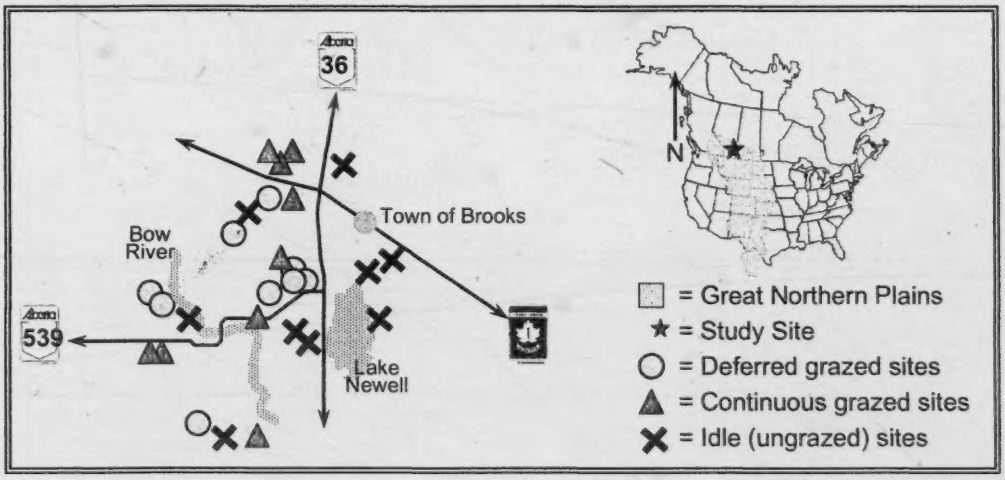
2.2 Objective

This chapter investigates the impact of three cattle grazing regimes on the water quality of prairie wetlands, and the subsequent ecological impacts on the larval odonate (Insecta: Odonata) fauna.

2.3 Study area

Study sites are located within an 1800 km^2 area of the dry mixed-grass prairie in south-eastern Alberta, Canada (50° 30' N, 111° 55' W) (Figure 2-2). Agriculture (i.e. cropland, irrigation infrastructure, beef cattle grazing) and oil and gas extraction are the main contemporary sources of disturbance within the study area. Free-roaming bison herds and wildfires were past sources of landscape disturbance, but these have been eliminated as 94% of Alberta's native grasslands have been converted to cropland (Environment Canada 2002). The dry mixed-grass prairie still supports substantial tracts of grazing land because precipitation is insufficient for dependable cropping regimes.

Figure 2–2: Study area location and distribution of study sites.



Moisture deficits are typical in this dry and windy region, particularly at the end of summer (i.e. 250mm mean annual precipitation; 19 km/h mean annual wind

speed) (Environment Canada 2002). Study site ponds are not typical of naturally occurring ephemeral prairie wetlands, however, because water levels are artificially controlled by the Eastern Irrigation District (EID) and Ducks Unlimited Canada (DUC) via a network of irrigation canals. These managed wetlands remain wet into late summer and have been created to serve as irrigation basins and sources of drinking water for beef cattle and to a lesser degree, to provide wildlife habitat.

2.4 Methods

2.4.1 Experimental design

Wetlands were selected based on five criteria: grazing regime during the previous five years, basin characteristics (i.e. size, source, and degree of isolation represented at least once in each treatment), surrounding habitat (i.e. grasslands only, no croplands), wetland class (i.e. Stewart and Kantrud (1971) Class IV or V), and access permission.

Manipulating pasture sizes and stocking rates for the sake of this project was logistically infeasible. Continuous and rotational grazing are the two predominant grazing strategies employed within the study area. Continuous grazing regimes keep cattle in one pasture all season (i.e. May to August) and consequently involve small herds of cattle on small sections of private land. More commonly, rotational grazing merges many herds to form a single 'super herd' managed by a local grazing association. To ensure pasture re-growth and a constant supply of forage, cattle are directed through a series of pastures beginning with early-germinating tame pastures (i.e. exotic species) and ending on native grasslands. Deferred grazing regimes are a

derivation of rotational grazing systems and are promoted by DUC in an attempt to decrease waterfowl nest exposure and mortality due to foliage removal and cattle trampling. DUC negotiates contracts with grazing managers to delay cattle access to specific wetlands until after July 15th, at which time most waterfowl broods have fledged (NAWMP 1999).

The experimental units for this study were the individual wetlands. Wetland characteristics were estimated from a continuous 250m stretch of shoreline that was selected to best represent the variation in wetland vegetation and shoreline structure. All data collection took place along these 250m segments (delineated into six 50m sections selected as random sub-samples) to standardize sampling wetlands of various sizes. The grazing regime in the pasture surrounding each wetland supported one of three treatments: deferred grazing (i.e. mid-July to mid-August), continuous grazing (i.e. approximately May to August), and idle (ungrazed) pastures (i.e. control treatment). There were six sampling rounds in 2000, each at 2-week intervals. Larval odonates were sampled using random stratified design (Krebs 1989); strata were delineated based on distance from interface of vegetation and open water. Pseudoreplication was avoided since experimental units (i.e. wetlands) were replicated nine times in each of the three treatments, and each wetland is far enough apart (i.e.>1km) to be considered statistically independent relative to dispersal distances (Hurlbert 1984). Although wetlands are connected via irrigation canals they are distinct habitats (i.e. flowing vs. standing water, sparsely vegetated banks vs. heavily vegetated shorelines), and canals are dry and unsuitable for aquatic organisms for a large portion of the year (i.e. late summer through spring); therefore wetlands

were considered biologically unconnected and statistically independent. Grazing was ubiquitous throughout the study area and as a result, the limitation on sample size was the number of ungrazed (idled) pastures in local protected areas. A total of twenty-seven wetlands were selected for a balanced design of nine replicates in each of the three treatments (Figure 2-2). Response variables included wetland water quality and larval odonate community metrics. Wetland vegetation structure, adult odonate community, and other aquatic invertebrates are addressed in following chapters.

2.4.2 Measuring wetland water chemistry

Basic water chemistry data were recorded with a handheld multi-meter (YSI Model 85 Dissolved Oxygen and Conductivity Meter) at each wetland for all rounds beginning with round three in 2000. Three of the six survey points were randomly selected for each sampling period and measurements were averaged to obtain a composite measurement for each site. Temperature (°C), pH, salinity (g/L, temperature corrected), and dissolved oxygen (%) were recorded at each survey point at 1m into the open water zone and at a depth of 15-25cm immediately upon arrival to avoid confounding effects from research activities that disturb wetland sediments.

2.4.3 Measuring water quality

Budget constraints dictated that the minimum number of water samples be submitted for analysis. All samples were collected three or more days after a storm event to minimize effects from run-off. Three of the six survey points were randomly selected, and 1L of water was collected from each point at 1m into the open water

zone at a depth of 30cm. The three samples were combined and one 0.5L composite sample was packed in ice and shipped to the lab that day.

Two water samples were collected from each wetland: 1) before grazing at deferred sites (i.e. mid-July 2000), herein called pre-cattle sample, and 2) during cattle grazing at deferred sites (i.e. late-August 2000), herein called post-cattle sample. These samples were analysed for ammonium (NH_4+), nitrates and nitrites (NO_2+NO_3), total Kjeldahl nitrogen (TKN), total phosphorus (TP), total dissolved phosphorus (TDP), total dissolved solids (TDS), and Chlorophyll-a concentrations (i.e. algae productivity) at the Limnology Lab at the University of Alberta. An additional sample was collected from all ponds at the end of the grazing season in late August 2000, and analyzed locally at Lakeside Packers (Brooks, AB) for fecal coliform concentrations.

2.4.4. Collecting larval odonates and detecting fish

During each sampling round, odonate larvae were collected along with other aquatic macro-invertebrates using a D-frame sweep net (one of the best devices for sampling aquatic invertebrates in heavily vegetated wetlands; Turner & Trexler 1997). Three of the six survey points were randomly selected, and three distinct micro-habitats were sampled at each point including: emergent vegetation (i.e. 1m outward from open water), emergent vegetation /open water interface, and deeper open water (i.e. 1m inward from vegetation/water interface). Five 1m sweeps were collected from each microhabitat for a total of forty-five sweeps per wetland. Larvae were preserved in 70% ethanol solution until later identified to genus (i.e. lowest

identifiable taxonomic level). Primary references for identification were Clifford (1991) and Merritt and Cummins (1996).

Fish were detected by randomly placing bread baited minnow traps at three of the six delineated survey points in the open water zone of each site for a two to three day period (checked daily) during the 2001 field season.

2.4.5 Statistical analysis

The Shapiro-Wilk test was used to check if data were normally distributed and Levene's Statistic was used to test the homogeneity of variances (SPSS 1999, Zar 1999). Repeated measures analyses of variance (or Friedman's tests for non-normal data) were performed to test for seasonal changes in water chemistry, water quality, and odonate fauna. I used Newman-Keuls *post hoc* multiple range tests to determine which means differed when significant differences were found with Friedman's tests. The Morisita-Horn index is a measure of the similarity in community composition between different communities, ranging from completely different to identical (i.e. score 0–1, respectively) that is relatively independent of sample size or diversity (Wolda 1981). This similarity index was calculated for the larval odonate fauna per round for each site, and analysed for significant differences between treatments (i.e. arcsine transformed, Friedman's tests). An alpha of 0.10 was selected *a priori* for treatment impacts on water quality and larval odonates due to the difficulties in obtaining a representative measurement for an attribute as inherently variable as water quality, and the limited number of water samples. An alpha of 0.05 was selected *a priori* for all other analyses.

2.5 Results

2.5.1 Wetland water chemistry and grazing treatment

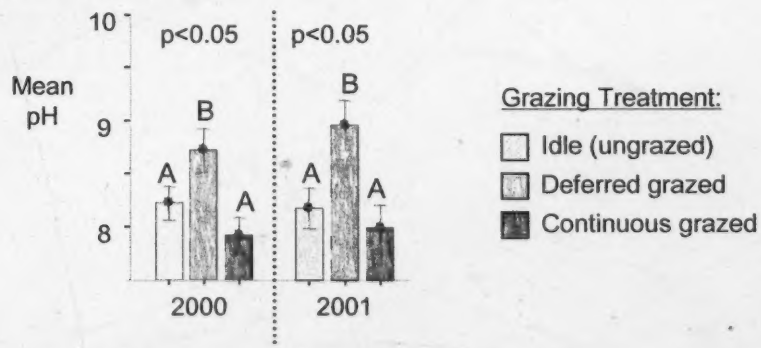
Wetland pH was significantly higher at deferred grazed compared to idle or continuously grazed sites in both 2000 and 2001, when considering sampling round (significant treatment effects, sampling period effects and interactions not significant; Table 2-1; Figure 2-3).

Salinity (g/L, temperature corrected) was significantly higher at wetlands with deferred grazed compared to continuously grazed regimes during 2001, considering round (significant treatment effects, sampling period effects and interactions not significant; no significant differences in 2000; Table 2-2; Figure 2-4).

Table 2-1: Results for differences in wetland pH using Friedman's test.
($?_{crit}^2$ values from Table B.1 in Zar 1999)

Year	Source	df	H	$?^2_{crit}$	p-value
2000	Treatment	2	8.829	5.991	<0.05
	Round	2	0.041	5.991	not significant
	Interaction	4	0.582	9.488	not significant
2001	Treatment	2	10.633	5.991	<0.05
	Round	2	5.537	5.991	not significant
	Interaction	4	<1	9.488	not significant

Figure 2-3: Wetland pH and grazing treatment.
(Bars represent ±1 SE)

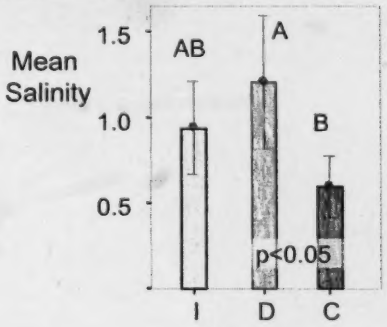


Grazing treatment did not significantly affect wetland temperature (p=0.154), or dissolved oxygen (p=0.258) in either year of data collection, considering sampling round (i.e. repeated measures ANOVA).

Table 2-2: Results for differences in wetland salinity using Friedman's test.
($?_{crit}^2$ values from Table B.1 in Zar 1999)

Year	Source	df	H	$?^2_{crit}$	p-value
2001	Treatment	2	6.338	5.991	<0.05
	Round	2	<1	5.991	not significant
	Interaction	4	<1	9.488	not significant

Figure 2-4: Wetland salinity and grazing treatments, 2001.
(Bars represent ±1 SE; I=idle, D=deferred, C=continuous)



2.5.2 Cattle impacts on wetland water quality

The impact of cattle urine on water quality as a result of cattle grazing at a wetland was measured via total dissolved solids (TDS). No significant differences in TDS were found between grazing treatments when considering time (Friedman's test).

Organic inputs from cattle feces in and around a grazed wetland were assessed by analysing differences in fecal coliform, nitrogen, phosphorus, and chlorophyll-a

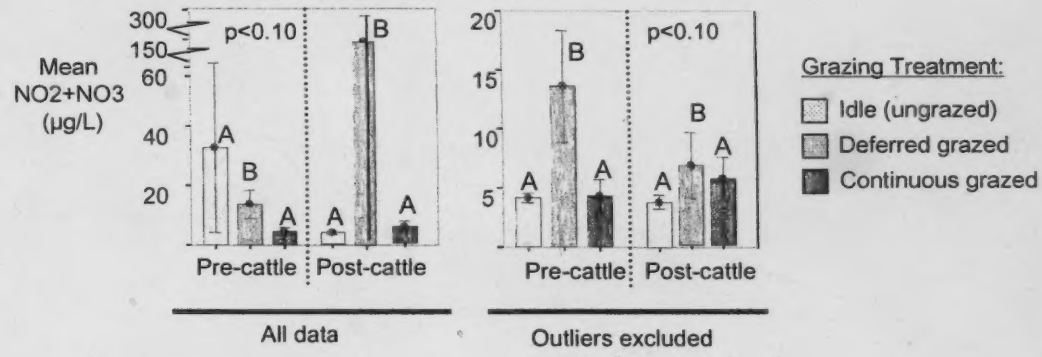
concentrations. No significant differences in fecal coliforms (p=0.861), chlorophyll-a, total dissolved phosphorus, total phosphorus, total Kjeldahl nitrogen (TKN), total nitrogen (i.e. NO_2+NO_3 + TKN), or ammonium were found between grazing treatments when considering time (Friedman's test for all); time was not significant for any water quality parameter.

Significantly higher concentrations in NO_2+NO_3 (µg/L) were present at deferred grazed sites, as compared to continuously grazed or idle sites, for both pre- and post-cattle samples (power=0.50; Table 2-3; Figure 2–5). However, even when outliers were omitted (i.e. one site from both idle and deferred grazed wetlands) to reduce the high level of within-treatment variation and maintain homoscedasticity (i.e. values that violated the assumption of equal variances were considered outliers and omitted), significantly higher NO_2+NO_3 concentrations were found only within the pre-cattle sample, and therefore can not be associated with cattle grazing (power=0.35; Table 2-3; Figure 2-5).

Table 2-3: Results for differences in wetland NO_2+NO_3 levels using Friedman's test.
($?_r^2$ values from Table B.1 in Zar 1999)

Outliers present	Source	df	H	$?^2_{crit}$	p-value
Yes	Treatment	2	6.402	4.605	<0.10
	Round	1	2.822	2.706	<0.10
	Interaction	2	2.764	4.605	not significant
No	Treatment	2	5.460	4.605	<0.10
	Round	1	3.035	2.706	<0.10
	Interaction	2	4.263	4.605	not significant

Figure 2-5: Wetland NO_2+NO_3 levels between grazing treatments.
(Bars represent ±1 SE)



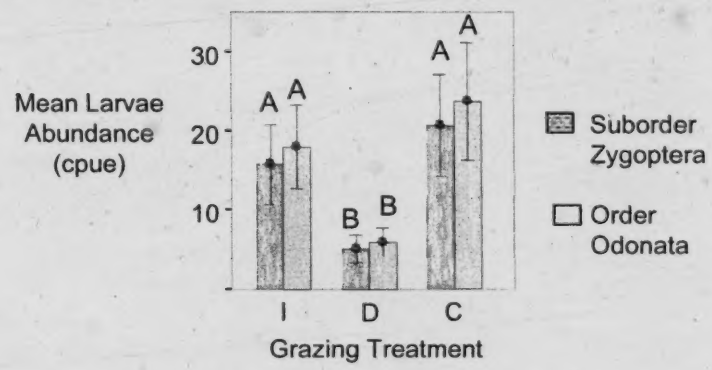
2.5.3 Cattle grazing and larval odonates

Five genera of larval odonates were collected from the study site including the dragonflies *Aeshna, Anax,* and *Sympetrum*, and the damselflies *Enallagma* and *Lestes*. Analysis was conducted by genus as well as by higher taxonomic groups (i.e. Suborder Anisoptera (dragonflies), Suborder Zygoptera (damselflies), and Order Odonata. Only data from 2000 were analysed to coincide with the year water quality samples were collected. No significant treatment effect was found for Anisoptera, Zygoptera, or overall genus richness, when considering sampling round (Friedman's test). Zygoptera and overall Odonata abundance was significantly lower at deferred grazed wetlands compared to idle or continuously grazed wetlands (significant treatment effects, sampling period effects and interaction not significant; Table 2-4; Figure 2-6). No treatment effect was found regarding differences in Anisoptera abundance.

Table 2-4: Results for differences in larval abundance using Friedman's test.
($?_r^2$ values from Table B.1 in Zar 1999)

Group	Source	df	H	$?^2_{crit}$	p-value
Zygoptera	Treatment	2	8.413	4.605	<0.10
	Round	5	5.853	9.236	not significant
	Interaction	10	4.625	15.987	not significant
Odonata	Treatment	2	6.560	4.605	<0.10
	Round	5	3.286	9.236	not significant
	Interaction	10	4.854	15.987	not significant

Figure 2-6: Larval odonate abundance among grazing treatments.
(Bars represent ±1 SE; I=idle, D=deferred, C=continuous)

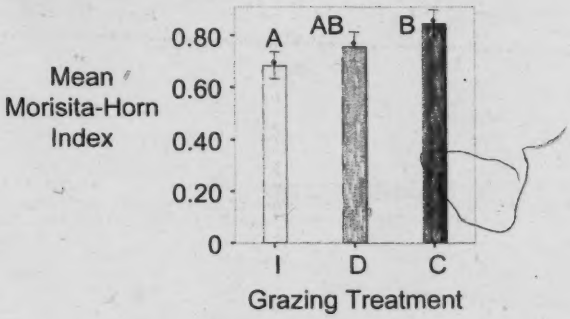


Significant differences in larval odonate composition were found between wetlands with idle and continuous grazing regimes, when considering sampling round (significant treatment effects, sampling period effects and interaction not significant; Table 2-5; Figure 2-7).

Table 2-5: Results for differences in larval odonate similarity using Friedman's test.
($?_r^2$ values from Table B.1 in Zar 1999)

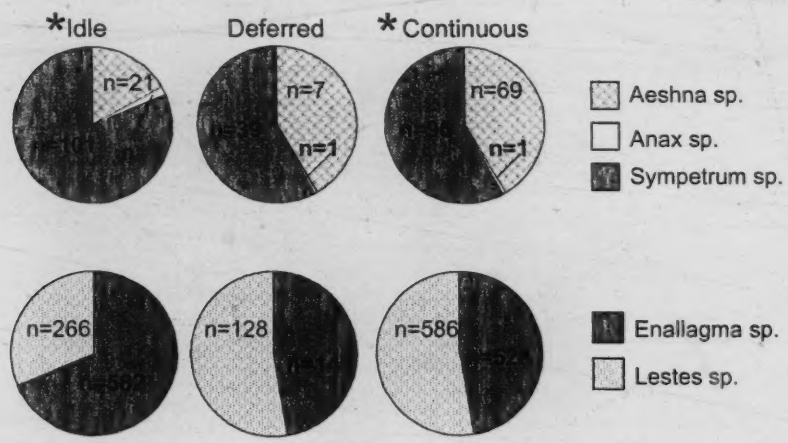
Source	df	H	$?^2_{crit}$	p-value
Treatment	2	7.620	4.605	<0.10
Round	5	7.803	9.236	not significant
Interaction	10	9.931	15.987	not significant

Figure 2-7: Differences in larval odonate genus similarity between treatments.
(Bars represent ±1 SE; I=idle, D=deferred, C=continuous; plots of untransformed data)



Closer examination of the genus composition of larval odonates between grazing treatments reveals differences in the relative abundance of both Anisoptera and Zygoptera larvae at idle and continuously grazed wetlands (Figure 2–8). *Sympetrum* spp. (5 possible species) and *Enallagma* spp. (5 possible species) dominate at ungrazed sites, but near equal numbers of *Sympetrum* spp. and *Aeshna* spp. (2 possible species), as well as *Enallagma* spp. and *Lestes* spp. (3 possible species) exist at continuously grazed sites.

Figure 2-8: Larval odonate composition and relative abundance among grazing treatments.
(* = significant differences)

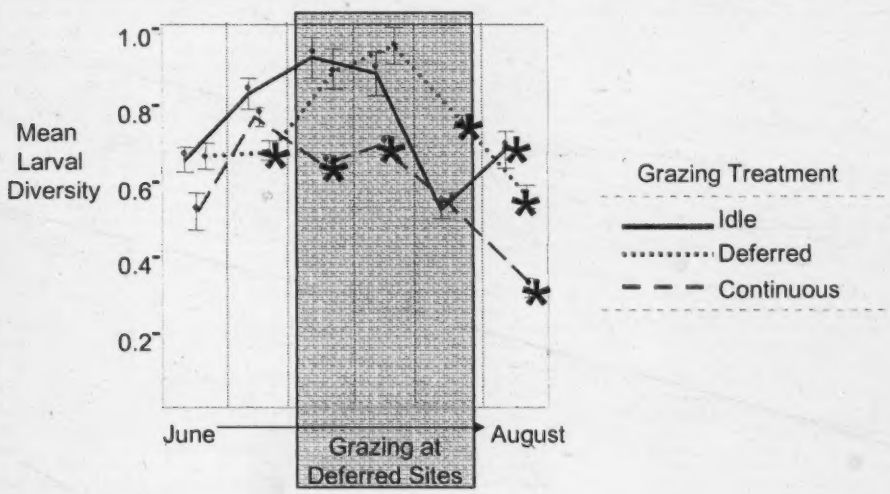


Shannon diversity indices measure the number and evenness of species in a community and were used to assess differences in larval odonate diversity between treatments. Both treatment and round were significant variables with respect to larval odonate diversity (significant treatment and sampling period effects, and significant interaction; Table 2-6; Figure 2–9). Overall larval odonate diversity tended to increase over summer, peaking in July. Larval diversity was significantly lower at continuously grazed wetlands compared to idle and deferred grazed wetlands during Round 2-6. Larval diversity at idle and deferred grazed sites was relatively comparable and high for rounds one to four. Larval odonate diversity was highest at deferred sites in round five (i.e. immediately after grazing stopped), and at idle sites in round six.

Table 2-6: Results for differences in larval odonate diversity using Friedman's test.
($?_r^2$ values from Table B.1 in Zar 1999)

Source	df	H	$?^2_{crit}$	p-value
Treatment	2	7.269	4.605	<0.10
Round	5	86.587	9.236	<0.10
Interaction	10	30.061	15.987	<0.10

Figure 2–9: Differences in larval odonate genus diversity at wetlands with different grazing treatments
(Bars represent ±1 SE, * = significant differences when a=0.10)



2.5.4 Fish presence

No significant differences in the presence/absence of fish at study wetlands were found between treatments (i.e. Kruskal-Wallis analysis for differences among treatments in the number of sites with fish; p=0.538). Fish were found at all but six of the twenty-seven study ponds. Brook stickleback (*Culaea inconstans*) were the only fish species detected in study wetlands.

2.6 Discussion

Wetland hydrology is the primary influence on wetland properties such as pH, oxygen content, and nutrient availability (Mitsch & Gosselink 2000). The hydrogeomorphology of a wetland refers to its hydrology (e.g. flooding period, water source), geomorphology (e.g. landscape topography, degree of isolation), and climate (Brinson 1993). Initial site selection attempted to select template wetlands whose hydrogeomorphology was as similar as possible, with cattle grazing regime being the only major difference among sites.

2.6.1 Wetland water chemistry

Basic wetland water chemistry data were collected to characterize study sites, as well as to compare treatments and account for any consequent effects on the odonate community. Dissolved oxygen fluctuates widely both spatially and temporally (e.g. daily) within wetlands, and therefore, was not a useful parameter for characterising study ponds. Redox potential (i.e. a quantitative measurement of the electron availability of a solution, and of the "...tendency of a soil to reduce or oxidize a substance" (Mitsch & Gosselink 1993)) is a more reliable quantitative measure for wetland oxygen (i.e. decreases in a predictable fashion as a sequence of reducing reactions occur) (Mitsch & Gosselink 2000), and is recommended in place of dissolved oxygen for future studies. Additionally, macroinvertebrates that respire via gills (i.e. odonates) are indicative of well-oxygenated wetlands since they can not survive long periods of anoxia (Murkin et al. 2000).

Wetland temperature fluctuates diurnally, but critical thresholds occur during the winter (not summer) months (Murkin et al. 2000). Treatments were compared to see if the removal of wetland vegetation via grazing affects water temperature. No significant difference was detected suggesting the presence of emergent vegetation has little impact on water temperature; however, the scale of measurement was not appropriate to detect micro-habitat changes. Water temperatures were recorded 1m into the open water zone, rather than within the emergent vegetation zone where cattle typically cluster (Jansen & Robertson 2001). Grazing may impact the micro-habitat within the littoral zone, but does not appear to affect overall wetland temperature.

Wetland pH is primarily determined by soil type; mineral soils are usually alkaline while organic soils are typically acidic (Mitsch & Gooselink 2000). The pH data for all study sites, including the significantly higher deferred grazed sites (i.e. $mean_{2000}$ =8.72, SD=1.02; $mean_{2001}$= 8.96, SD=1.22), are well within the expected range of pH 6-10 for prairie wetlands and likely not biologically significant to aquatic organisms (Mitsch & Gosselink 1993). A slight increase in pH could alter the availability of wetland nutrients by increasing their solubility (National Science Foundation 2003). The link between higher pH and intensive cattle use (i.e. deferred grazed sites) is most likely the wetland soil; trampling of the shoreline by grazing/drinking cattle may disturb wetland sediments and introduce more of the alkaline parental material to the water, thereby increasing wetland pH.

Salinity levels range from nearly fresh to near sea water concentrations on the prairies and are primarily determined by the interaction of groundwater,

evapotranspiration, and soil type (Mitsch & Gosselink 2000). Although statistically higher salinity was detected at deferred grazed sites in 2001, these wetlands are relatively fresh (i.e. $mean_{deferred\ 2001}$=1.20g/L, SD=2.02; $mean_{others\ 2001}$=0.899g/L, SD=1.54). It is unlikely this increase is biologically significant since salinity levels are well within known tolerance limits of aquatic invertebrates (i.e. most sensitive species thrive in waters 0-5g/L), and within the salinity range required for most wetland vegetation (Murkin et al. 2000).

2.6.2 Wetland water quality

Cattle tend to concentrate around prairie wetlands and loiter in the water while they drink and graze, as observed in this study and others (Nader et al. 1998, Jansen & Robertson 2001). Wetland water samples were analysed for substances present in cattle excrement. Nitrogen (i.e. TKN, NO_2+NO_3, NH_4+), phosphorus (i.e. TP and TDP), and fecal coliforms are found in cattle manure, while the ions in TDS are found in cattle urine (pers. comm. Richard Casey 2000). Chlorophyll-a was measured to test for differences in wetland productivity with respect to algae as a result of increased nutrients.

Significantly higher nitrate and nitrite (NO_2+NO_3) concentrations were detected within the deferred grazing treatment (Figure 2-5), however these elevated nitrogen levels can not be attributed to cattle since they were recorded before cattle began grazing these sites (i.e. prior to July 15th). Differences in NO_2+NO_3 are not likely due to management activities prior to the study since NO_2NO_3 is quickly assimilated by plants, lost in groundwater, or transformed via denitrification (Mitsch

& Gosselink 1993). Nitrogen differences cannot be attributed to dissimilarities in nitrogen fixing blue-green algae concentrations since chlorophyll-a data (i.e. index for algae) between treatments were similar. It is probable that the higher pH of deferred grazed sites (Figure 2-3) is affecting NO_2+NO_3 concentrations by increasing nutrient solubility (National Science Foundation 2003). Sources of NO_2+NO_3 other than livestock manure include rainfall, agricultural fertilizers, and decomposing organic matter (Mitsch & Gosselink 2000), but it is unlikely that rainfall or agricultural pollutants are driving this difference in NO_2NO_3 since all study sites are influenced by the same general weather pattern, and wetlands surrounded by croplands were not selected for study. Higher pH may increase the rate of organic matter decomposition (Mitsch & Gosselink 2000) and thereby indirectly increase wetland NO_2+NO_3 concentrations. Toxicity levels for NO_2+NO_3 are at least two orders of magnitude larger than the maximum concentrations recorded in this study (i.e. 2 500 – 100 000µg/L for amphibians and >100 000µg/L for cattle and humans), therefore these elevated levels likely have little impact on vertebrate wetland biota. Little is known regarding the ecological impact of nutrient loading on aquatic invertebrates in freshwater wetlands; however preliminary research shows a positive relationship between nutrient loading and overall aquatic invertebrate abundance due to an increase in overall wetland productivity (Murkin et al 1991, Campeau et al. 1994). The seasonal drop in NO_2+NO_3 concentrations due to plant uptake in early summer may explain the lack of a significant difference within the second (post-cattle) sample (Bender 1976).

It is unclear whether the lack of significant difference in water quality between treatments is due to the high buffering capacity of prairie wetlands, or the inherently variable nature of water quality data exacerbated by an insufficient sampling regime; however the former is most likely. Prairie wetlands are known to function as nitrogen and phosphorus sinks, and consequently nutrient-rich in-flows have minimal effects on the concentration of available nutrients and no impact of long-term nutrient budgets (Mitsch & Gosselink 2000). It is likely that the level of disturbance and nutrient inputs from unrestricted cattle grazing is too low to biologically impact prairie wetlands, which have evolved with, and are dependent on, disturbance (i.e. periodic drought and grazing from bison herds). Conversely, environmental data alone may be insufficient to properly make this assessment. Nutrient inputs to nitrogen-limited systems (i.e. prairie wetlands) will be quickly absorbed by wetland vegetation (Mitsch and Gosselink 1993) and will be difficult to detect via environmental data. Cattle activity (i.e. defecation, trampling) at the wetland shore is an inherently intermittent event, and it is therefore unfeasible to sample water immediately after a disturbance event to measure maximum impact. Sampling efforts were conservative and may have been too infrequent to detect the impact of an intermittent disturbance such as cattle defecation, urination, and trampling (i.e. 50% chance of committing Type II error with outliers included, 35% chance with outliers excluded). Analysis of the impact of grazing treatment on wetland biota (i.e. adult odonates and other aquatic macro-invertebrates) is required before the effect of cattle grazing on wetland habitat quality can be fully assessed.

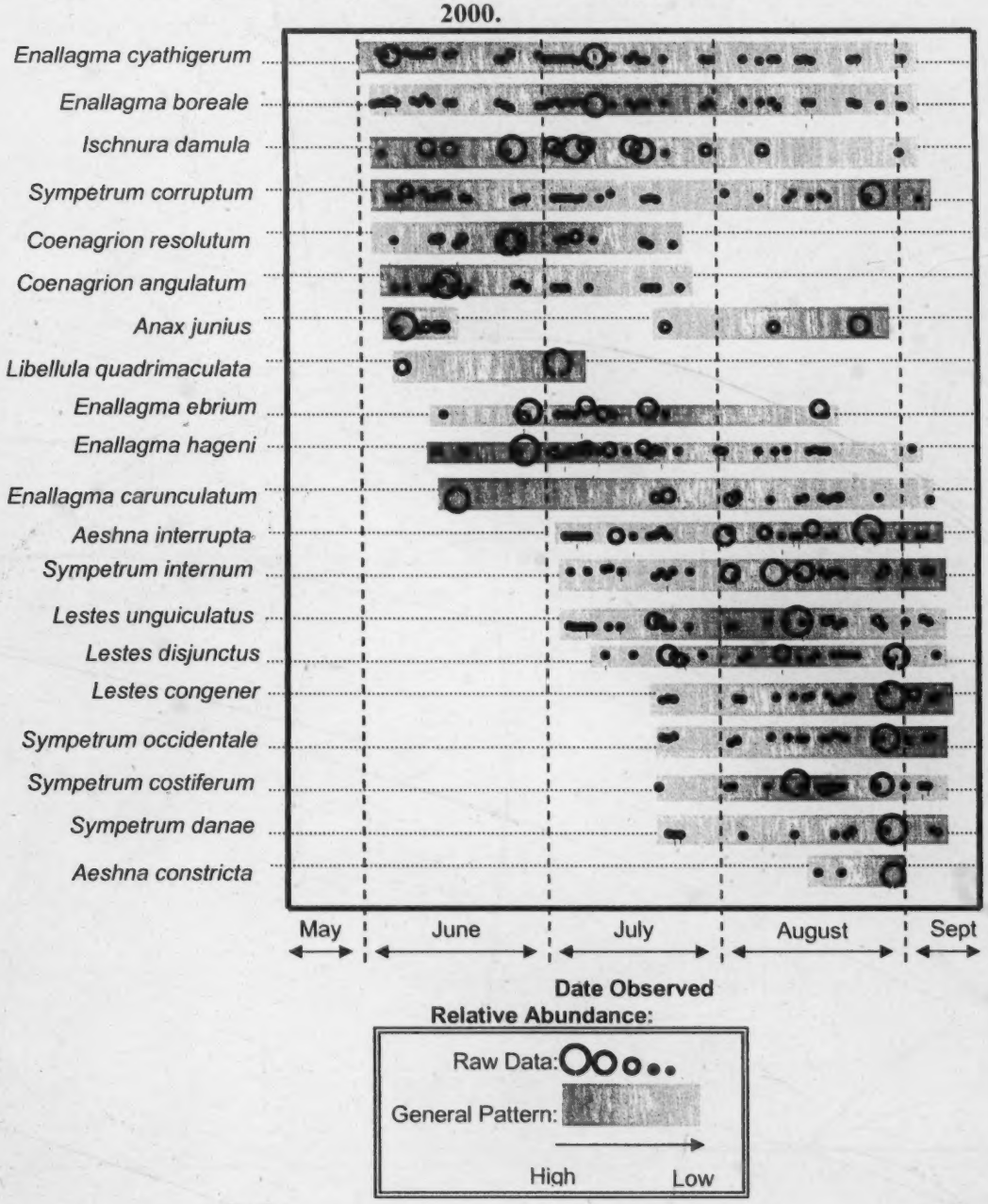
2.6.3 Larvae and grazing treatment

Odonates are predacious macro-invertebrates that occupy both aquatic and terrestrial habitats at different stages of their life cycle. Long-lived aquatic larvae require unpolluted, oxygenated, fresh water, submerged vegetation as cover for avoiding predation and detection by prey, emergent vegetation as emergence structures, and other aquatic invertebrates as prey (Corbet 1999). Terrestrial adults concentrate around water bodies and depend on aquatic vegetation as territorial perches, oviposition cues, and shelter from winds (Corbet 1999). Different species emerge and mature throughout the season causing a shift in adult species composition throughout the season (Figure 2–10).

Brook Stickleback (*Culaea inconstans*) and their consequent predatory pressure on larval odonates are essentially ubiquitous, present at wetlands regardless of treatment, and likely not responsible for any differences in larval odonate communities. Larval odonates eat small Brook Stickleback fry, and conversely adult Brook Stickleback have a small gape size capable of eating small (i.e. early instar) odonate larvae.

Surveying wetland biota, unlike environmental parameters, incorporates time since the presence and condition of relatively long-lived organisms reflects past events (i.e. intermittent pollution) rather than simply conditions at the time of measurement (Rosenburg and Resh 1996). Larval odonate communities were analysed to test for significant differences among treatments that may not have been statistically attributable to any of the specific environmental parameters measured in this study, reflecting antecedent conditions instead.

Figure 2–10: Seasonal changes in adult odonate composition and abundance, 2000.



Significantly fewer larvae were collected at deferred grazed wetlands rather than continuously grazed wetlands, as initially predicted (Figure 2–6). Deferred grazing regimes involve combining many herds of cattle into one "super herd" (i.e. hundreds of cattle) whereas continuously grazed wetlands involve smaller grazing operations. The number of cattle rather than simply the duration of their presence may be more important than initially recognized, making deferred grazing a more acute grazing disturbance. This difference is largely reflected in the Suborder Zygoptera due a larger sample size since seven times more Zygoptera vs. Anisoptera larvae were collected (i.e. mean $_{\text{Total Zygoptera}}$=2235, mean $_{\text{Total Anisoptera}}$=337). This relative abundance was also observed in adult odonates as adult damselflies were observed to be far more abundant in the field than adult dragonflies, especially with respect to the family Aeshnidae.

Habitat quality must be evaluated in terms of survival and reproductive success in concert with abundance data (van Horne 1983). Although the abundance of odonate larvae at idle and continuously grazed wetlands is similar, the proportion of larvae that successfully emerge and reproduce is unknown. Odonates are very vulnerable to physical disturbance during emergence (Corbet 1999), and presumably cattle activity along shorelines (i.e. walking through vegetation) will disturb emerging odonates and decrease emergence success, making continuously grazed habitat more hostile environments for odonates than ungrazed sites, despite similar numbers of larvae.

Deferred grazing is associated with decreased abundance of odonate larvae at wetlands. The littoral zone is the link between cows and odonates; this is where most

odonate larvae exist and where most of the physical disturbance from foraging and loitering cattle occurs (i.e. cows were observed trampling and defecating on shorelines while browsing submergent and emergent vegetation). Analysis of water samples found no link between grazing and water quality, however significantly lower larvae abundances suggests that large numbers of cattle (i.e. deferred grazed sites) compromise littoral habitat enough to impact the odonate community of a wetland. Larval odonate fauna represent previous adult odonate fauna via generational lag and therefore fewer larvae may reflect a decrease in ovipositing adults, which cue into wetland vegetation for ovipositing sites (Corbet 1999). Figure 2–10 shows that the majority of the mature odonate fauna at the time of deferred grazing (i.e. mid-July to early-August) are damselflies of the genus *Enallagma*, which is consistent with the decline in damselfly larvae.

The larval odonate community of each treatment is compared using the Morisita-Horn similarity index which incorporates presence/absence, abundance, and genus richness, and ranges from 0 to 1 (i.e. 1 most similar). Taxonomic limitations make identification of *Coenagrion* spp., *Ischnura* app., and *Libellula* spp. larvae very difficult. High numbers of *Coenagrion* spp. and *Ischnura* spp. adults were detected, and the lack of positive identification of larvae in study wetlands is most likely due to taxonomic limitations and misidentification of these individuals as *Enallagma* spp. larvae. Larvae identification were maintained at the genus level rather than the family level to make richness and diversity estimates more precise. Grouping three genera into one class due to taxonomic limitations will have the effect of making it harder to detect significant differences in larvae communities among study wetlands,

yet differences were detected in this study. Larval odonate communities were significantly less similar among idle (ungrazed) compared to continuously grazed wetlands (Figure 2–7). There are relatively more *Enallagma spp.* and *Sympetrum spp.* and less *Lestes spp.* and *Aeshna spp.* larvae at idle compared to continuously grazed wetlands (Figure 2–8). No significant difference in wetland water quality was detected between grazing treatments to explain this shift in larval odonate composition. Larvae from the two Zygoptera genera have similar foraging behaviours (i.e. they crawl amongst submerged vegetation as larvae, and glean vegetation as adults) and ovipositing strategies (i.e. endophytic) but differ in over-wintering strategies (Corbet 1999). Over-wintering strategies are more related to wetland hydrology than habitat quality (i.e. *Lestes spp.* oviposit inside vegetation above the water line and over- winter as diapause eggs as an adaptation to ephemeral prairie wetlands; Corbet 1999), and do not explain differences in larval composition since all wetland water levels are similarly controlled.

Both Anisoptera genera over-winter as diapause eggs and depend on wetland vegetation during ovipositing (i.e. all *Aeshna spp.* and most *Sympetrum spp.*). *Aeshna spp.* larvae are 'climbers' that crawl along submerged vegetation and other substrates, whereas larval *Sympetrum spp.* are considered 'sprawlers' that crawl along wetland sediments (Corbet 1999). More submersed vegetation exists at continuously grazed sites and cattle chronically trample wetland sediments in the littoral zone, therefore conditions at continuously grazed sites may favour the climbers *Aeshna spp.* larvae over *Sympetrum spp.* larvae (Buskirk and Sherman 1985). Larvae odonate populations reflect previous adult odonate populations via generational lag and

therefore indicate at least minimal prior habitat suitability for adult odonates. *Aeshna spp.* adults are 'flyers' that hunt and defend territories on the wing, while *Sympetrum spp.* adults are 'perchers' and use riparian vegetation as a perch from which they hunt or defend (Corbet 1999). When chronic grazing decreases wetland vegetation structure (i.e. height; refer to Chapter 3) then there are fewer perches and therefore less suitable habitat for *Sympetrum spp.*, resulting in lower occurrence of oviposition by *Sympetrum spp.* at that wetland.

The Shannon diversity index is perhaps the most widely accepted diversity index since it incorporates evenness and is relatively independent of sample size (Wilhm and Dorris 1968). The seasonal pattern of diversity observed in larval odonates in Figure 2–9 (peaked in July) was also observed in adult odonates throughout the study area (Figure 2–10). Continuously grazed wetlands consistently had significantly lower larval odonate diversity, whereas the larval diversity between deferred and idle sites remains similar until the end of the summer. Once cattle were removed from deferred grazed pastures the larval odonate diversity in those wetlands was significantly higher than at wetlands with any other grazing treatment. These results are consistent with the Intermediate Disturbance Hypothesis (IDH) which suggests both extreme and trivial disturbance decrease diversity (Connell 1978). The chronically disturbed continuously grazed sites represent sufficient disturbance to suppress odonate diversity (Figure 2–10). Idle wetlands represent minimal disturbance according to the IDH since idle and ungrazed deferred sites (i.e. pre-July 15th) are similar until grazing ended in late July, at which time odonate diversity temporarily increases at deferred sites. Similar patterns in odonate diversity with

respect to habitat disturbance have been found in lotic habitats (Stewart & Samways 1998).

Significantly higher community similarity and lower diversity indices at continuously grazed sites suggest that these sites are inhabited by generalist species or adaptive species that can withstand chronic disturbance. Taxonomic limitations restrict the identification of larval odonates to the species level however adult odonates are easily identified to species and therefore analysis of adult odonate data is needed to further support this hypothesis (see Chapter 3 and 4 this thesis).

2.7 Summary and recommendations

Water chemistry data support that all twenty-seven study wetlands share similar hydrogeomorphology and differ predominantly according to grazing regime. No significant difference in water quality due to cattle presence was detected among grazing treatments, and therefore I reject the hypothesis that nutrient inputs from cattle excrement measurably impacts wetland water quality (i.e. falsify Hypothesis #1 and consequently invalidate Hypothesis #2). It is important to note that the cattle stocking rates are uncharacteristically low within the study area compared to other grazed regions in Alberta due to the small amount of precipitation and low carrying capacity of the pastures. Higher stocking rates may have a greater influence on wetland water quality. Furthermore, the opportunity exists for future research to study the impact of other non-point source agricultural pollutants such as pesticide or fertilizer run-off at wetlands greatly influenced by cropland.

Deferred grazing can be a more substantial disturbance than initially thought due to large number of cattle on these pastures. Fewer odonate larvae inhabit deferred grazed wetlands, presumably due to the acute disturbance resulting from hundreds of cattle focusing their grazing efforts on the littoral zone of one wetland (i.e. trampling and vegetation removal). Deferred grazing may be conceptualized as a brief bottleneck treatment. Although odonate larvae are more abundant at continuously grazed sites, the odonate communities at these wetlands are significantly less diverse than communities at either idle or deferred grazed wetlands. Further analysis involving vegetation and adult odonate fauna (identifiable to species rather than genera level; presented in Chapters 3 and 4) will help clarify the sensitivity of odonates to cattle grazing at prairie wetlands, and their subsequent suitability as bioindicators of this disturbance.

Grazing treatments were not contrived for this research but are current management regimes employed on the prairies of Alberta. For the purposes of this study, grazing was simplistically measured based on its duration (i.e. all summer, mid-summer, or not at all), but future research should be more sensitive to its complexity and the myriad opportunities associated with grazing impacts and timing. On-site livestock watering technology using renewable energy (i.e. solar powered water pumps) is available and offers wetland managers an option other than simply unlimited or prohibited cattle access. Grazing duration, timing (i.e. early, mid-, or late summer), frequency (i.e. annually or less frequent), and intensity (i.e. large vs. small herds) are all important synergistic variables that have not been addressed with respect to their impact on wetland flora and fauna.

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Chapter 3

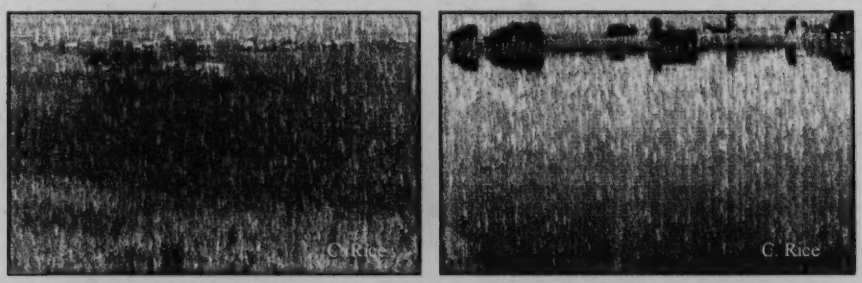
Adult odonates as bioindicators of cattle grazing and vegetation community at prairie wetlands

3.1 Introduction

The extensive use of wetlands as a grazing resource has persisted with little attention to the ramifications of grazing to riparian invertebrates. Agriculture dominates the Alberta prairie landscape and grazing is more common than cultivation in dry regions such as the dry mixed-grass prairie. Rotational grazing is the conventional management system and involves moving herds of cattle among a series of pastures throughout the summer. The uncommon alternative is grazing a single pasture all summer long at a lower stocking rate to ensure sufficient forage. Ungrazed (idle) wetlands are very rare in the modern prairie landscape.

Cattle in the study area (and throughout southern Alberta) have unrestricted access to wetlands occurring in pastures. On the dry mixed-grass prairie, cattle target their grazing efforts on the lush oasis of wetland vegetation and spend 5-20 times more time grazing riparian areas, lingering in the water to drink and graze submergent and emergent vegetation (Fitch & Adams 1998; Figure 3-1).

Figure 3-1: Cattle focussing grazing efforts on lush wetland vegetation.



Vegetation is integral to basic wetland functions such as filtering and slowing run-off, buffering potentially polluting nutrients, and increasing overall wetland productivity (Mitsch & Gosselink 1993). Basic ecosystem attributes affected by cattle grazing include: changed plant species composition (i.e. decreased richness, and vegetation density); disrupted ecosystem function (i.e. nutrient cycling, succession); and changed ecosystem structure (i.e. vegetation height, soil erosion) (Fleischner 1994). This study addresses the influence of cattle grazing on the composition and structure of prairie wetland vegetation and the subsequent ecological impacts on the adult odonate (Insecta: Odonata) fauna.

Vegetation provides critical habitat for odonates during the aquatic (larval), terrestrial (adult), and emergence phases of their life cycle. Riparian vegetation provides adult odonates with perch platforms, cover from predators, habitat cues for oviposition sites, foraging habitat for gleaning Zygopterans, and refugia from wind, which may be particularly important in windy locations such as the prairies of Alberta (Stewart & Samways 1998, Corbet 1999). Aquatic macrophytes reduce the risk of predation to larval odonates and decrease detection by potential predators, while emergent vegetation provides a platform for odonate emergence (Corbet 1999).

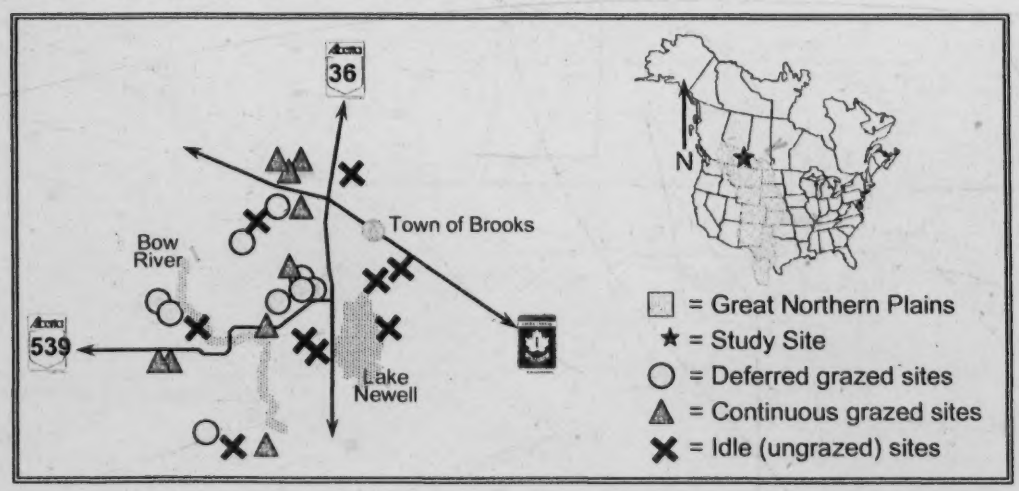
3.2 Objective

This research investigates the impact of three cattle grazing regimes on the vegetation communities of prairie wetlands, and the subsequent ecological impacts on the adult odonate (Insecta: Odonata) fauna.

3.3 Study area

Study sites are located within an 1800 km^2 area of the dry mixed-grass prairie in south-eastern Alberta, Canada (50° 30' N, 111° 55' W) (Figure 3-2). Agriculture (i.e. cropland, irrigation infrastructure, beef cattle grazing) and oil and gas extraction are the main contemporary sources of disturbance within the study area. Free-roaming bison herds and wildfires were past sources of landscape disturbance, but these have been eliminated as 94% of Alberta's native grasslands have been converted to cropland (Environment Canada 2002). The dry mixed-grass prairie still supports substantial tracts of grazing land since precipitation is insufficient for dependable cropping regimes.

Figure 3-2: Study area location and distribution of study sites.



Moisture deficits are typical in this dry and windy region, particularly at the end of summer (i.e. 250mm mean annual precipitation; 19 km/h mean annual wind speed) (Environment Canada 2002). Prairie wetlands are highly variable and many

dry up by the end of summer. Inter-annual variation is great also and follows a 5-20 year drought cycle (van der Valk & Davis 1978). The wetlands in this study however are intensively managed and are not typical ephemeral potholes. Wetlands are managed to serve as irrigation basins or waterfowl/shorebird habitat by the Eastern Irrigation District (EID) and Ducks Unlimited Canada (DUC). Water levels are intensively managed and kept unnaturally high and stable (i.e. wet all season). A consequence of decreased hydrologic variation has been a change in the typical vegetation community of these sites from a Class I/II/III (i.e. ephemeral, temporary, and seasonal ponds) to Class IV/V (i.e. semi-permanent and permanent ponds), and a subsequent increase in vegetation diversity (Stewart and Kantrud 1971).

3.4 Methods

3.4.1 Experimental design

Wetlands were selected based on five criteria: grazing regime during the previous five years, basin characteristics (i.e. size, source, and degree of isolation represented at least once in each treatment), surrounding habitat (i.e. grasslands only, no croplands), wetland class (i.e. Stewart and Kantrud (1971) Class IV or V), and access permission.

Manipulating pasture sizes and stocking rates for the sake of this project was logistically infeasible. Continuous and rotational grazing are the two predominant grazing strategies employed within the study area. Continuous grazing regimes keep cattle in one pasture all season (i.e. May to August) and consequently involve small herds of cattle on small sections of private land. More commonly, rotational grazing

merges many herds to form a single 'super herd' managed by a local grazing association. To ensure pasture re-growth and a constant supply of forage, cattle are directed through a series of pastures beginning with early-germinating tame pastures (i.e. exotic species) and ending on native grasslands. Deferred grazing regimes are a derivation of rotational grazing systems and are promoted by DUC in an attempt to decrease waterfowl nest exposure and mortality due to foliage removal and cattle trampling. DUC negotiates contracts with grazing managers to delay cattle access to specific wetlands until after July 15th, at which time most waterfowl broods have fledged (NAWMP 1999).

The experimental units for this study were the individual wetlands. Wetland characteristics were estimated from a continuous 250m stretch of shoreline that was selected to best represent the variation in wetland vegetation and shoreline structure. All data collection took place along these 250m segments (delineated into six 50m sections selected as random sub-samples) to standardize sampling wetlands of various sizes. The grazing regime in the pasture surrounding each wetland supported one of three treatments: deferred grazing (i.e. mid-July to mid-August), continuous grazing (i.e. approximately May to August), and idle (ungrazed) pastures (i.e. control treatment). There were six sampling rounds in 2000, each at 2-week intervals. Stratified random design was used for both adult odonate and vegetation samples (Krebs 1989); strata were delineated according to wetland vegetation zones as in Stewart and Kantrud (1971). Pseudoreplication was avoided since experimental units (i.e. wetlands) were replicated nine times in each of the three treatments, and each wetland is far enough apart (i.e.>1km) to be considered statistically independent

relative to dispersal distances (Hurlbert 1984). Although wetlands are connected via irrigation canals they are distinct habitats (i.e. flowing vs. standing water, sparsely vegetated banks vs. heavily vegetated shorelines), and canals are dry and unsuitable for aquatic organisms for a large portion of the year (i.e. late summer through spring); therefore wetlands were considered biologically unconnected and statistically independent. Grazing was ubiquitous throughout the study area and as a result, the limitation on sample size was the number of ungrazed (idled) pastures in local protected areas. A total of twenty-seven wetlands were selected for a balanced design of nine replicates in each of the three treatments (Figure 3-2). Response variables included wetland vegetation structure and adult odonate community metrics. Water quality, larval odonates and other aquatic invertebrates are addressed in other chapters.

3.4.2 Measuring the vegetation community

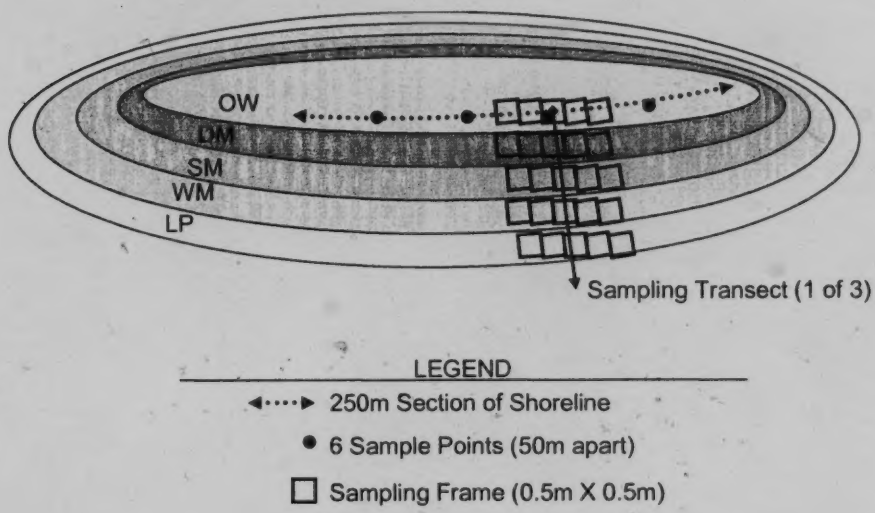
Vegetation surveys were conducted once per site at the end of summer in 2000 when the majority of plants had matured and were more easily identified. Grazing estimates were conservative for deferred sites because surveys occurred after grazing ceased, allowing for about two weeks of re-growth, whereas, at continuously grazed sites grazing was usually ongoing during surveys.

I measured vegetation characteristics relevant to the life histories of odonates including structure (i.e. height, abundance) and composition. Percent cover was estimated as a measure of plant abundance since it is the most common metric for estimating the percent of ground surface covered by the projection of a plant

(NARSC 1996). I used a Robel pole to measure plant height since it is the simplest and most accurate method for integrating both vegetation structure and density in prairie habitats (NARSC 1996, Ganguli et al. 2000). Grazing intensity is most often measured as percent utilization of palatable forage species, but I chose percent stems grazed for each plant species as a quantitative index of grazing intensity since I was interested in the impact of cattle grazing on the entire vegetation community rather than select palatable species (Holechek et al. 2001). I used sampling frames to measure percent cover, percent grazed stems, and vegetation composition (NARSC 1996), and chose a $0.25m^2$ quadrat (i.e. 0.5m by 0.5m) for this study since survey lines included both tall/dense and short/sparse vegetation (Daubenmire 1959). Most plants were identified to species except for willows and some non-flowering forbs (see Appendix-B for complete list).

The wetland vegetation community was composed of different zones of vegetation in concentric rings outward from the water's edge, influenced by the degree of water permanency (Stewart & Kantrud 1971). Due to the inherent decrease in vegetation height and abundance as distance from water increased (i.e. tall vigorous emergent vs. short sparse upland plants), comparisons between treatments were made within each of the five vegetation zones defined by Stewart and Kantrud (1971), including: open water, deep marsh, shallow marsh, wet meadow, and low-prairie (Figure3-3).

Figure 3-3: Vegetation survey design.
(OW=open water, DM=deep marsh, SM=shallow marsh, WM=wet meadow, LP=low-prairie)



Three of the six permanent survey points at each wetland were randomly chosen for each sampling round as starting points for vegetation transects. Rope transects were laid out perpendicular to the shoreline encompassing a span from 2m inside the open water zone outward to the beginning of the upland prairie zone, thereby traversing all wetland communities (Figure 3-3). Five sub-samples of quadrat and Robel pole measurements were taken from the middle of each vegetation zone, resulting in about 75 sub-samples per wetland.

3.4.3 Surveying adult odonates

Wetlands were surveyed at two-week intervals during the summer of 2000 (six sampling periods) to account for seasonal changes in odonate fauna. Odonates display diurnal movement patterns by roosting in upland vegetation at night and

concentrating around water bodies during the day. Diurnal patterns may be predicted at an even finer scale since mature males concentrate around the water from morning to afternoon, females make shorter trips to and from the water throughout the day, and immature males generally avoid riparian habitat until late in the afternoon when the density of mature males decreases (Moore 1953). Survey biases from the diurnal movement of odonates were avoided by randomizing the time of day at which surveys were conducted at each site and each sampling period.

Standardized survey methodology for adult odonates involved direct counts made while walking through riparian habitat during suitable flight conditions (i.e. low winds, warm, sunny). I used a hand-net to catch, identify, and release odonates (Moore 1953). Disturbance by the surveyor caused a flight-response making any sedentary odonates visible and thereby facilitating identification (Moore 1953). Each wetland vegetation zone was searched once in a serpentine-transect fashion. Abundance estimates were standardized based on time spent searching (i.e. initially 1½ hours per site, reduced to 45 minutes in 2000) and catch-per-unit-effort (CPUE) was calculated as the number of individuals counted/time spent searching.

Direct counts are estimates of the number of odonates at a 250m section of wetland. Despite a tremendous reservoir of individual odonates surrounding riparian habitats, odonate density in riparian/breeding habitat has been shown to remain fairly consistent due to territorial behaviour (Moore 1953). Direct counts are considered a conservative measurement of odonate abundance (Conrad et al. 1999).

In addition to abundance and richness, a reproductive effort index was developed to assess the impact of cattle grazing on odonate breeding success. The

reproductive index is based on similar indices developed for ornithology studies, where a range of observed breeding evidence/behaviours are used to weight abundance data according to certainty of reproductive success (Table 3-1).

Table 3-1: Abundance weights for odonates observed at time of vegetation surveys.

Rank	Observation
1	Mature male observed at site for 2+ consecutive rounds
2	Tandem pairs and/or females observed ovipositing
3	Very teneral individuals observed at site
4*	Exuvia collected*

*Exuvia were not collected in this study, but are included in the table to emphasize their importance in identifying breeding sites for odonates, and to promote their use in future studies.

The index is based on observations of mature male odonates, which are therefore weighted "1" to avoid artificially inflating the index (Schmiegelow et al. 1997). Only males of a certain species that were observed at a site for more than two rounds were included in the index to differentiate resident species from probable drifting species (Vickery et al. 1992). Ranks were calculated according to observations of reproductive behaviour per individuals of a species, rather than per site. Ranks are not additive.

3.4.4 Relating vegetation and odonate data

Vegetation characteristics significantly affected by grazing were related to odonate data using linear regression to examine what influence, if any, cattle-altered vegetation has on adult odonates. Only species with flight seasons corresponding to

the timing of vegetation surveys were included in the analysis (i.e. *Libellula quadrimaculata, Coenagrion resolutum,* and *C. angulatum* excluded, Figure3-4). Odonates were analysed according to Order Odonata, Suborder Anisoptera, Suborder Zygoptera, and individual species, as well as by three foraging groups: 'Flyers' (i.e. large dragonflies that catch prey in flight; includes Aeshnidae, *Libellula spp.*), 'Perchers' (i.e. medium dragonflies that dart out from perches; includes Genus *Sympetrum*), and 'Gleaners' (i.e. glean prey off plants and includes Suborder Zygoptera) (Corbet 1999, Needham et al. 2000).

3.4.5 Statistical analysis

Prior to analysis normal and Poisson distributions were tested using Shapiro-Wilk and Kolmogorov-Smirnov tests, and homogeneity of variances was tested for using the Levene statistic (SPSS 1999, Zar 1999). An *a priori* alpha of 0.05 was selected for all statistical tests. Where parametric assumptions could not be met, Kruskal-Wallis analyses were used to test for differences in vegetation parameters (i.e. height, %cover, %stems grazed, diversity and composition) and adult odonate parameters (i.e. abundance, richness, reproductive effort, diversity, and composition) between grazing treatments. All proportion (i.e. Morisita-Horn indices) and percent data (i.e. %cover, % stems grazed) were arcsine transformed before analysis to account for truncated distributions. Tukey's (parametric) or Nemenyi's (non-parametric) *post hoc* comparisons were performed to identify which treatments were significantly different. Friedmans's test, a nonparametric repeated measures procedure, was used to test for differences in odonate diversity and compositional

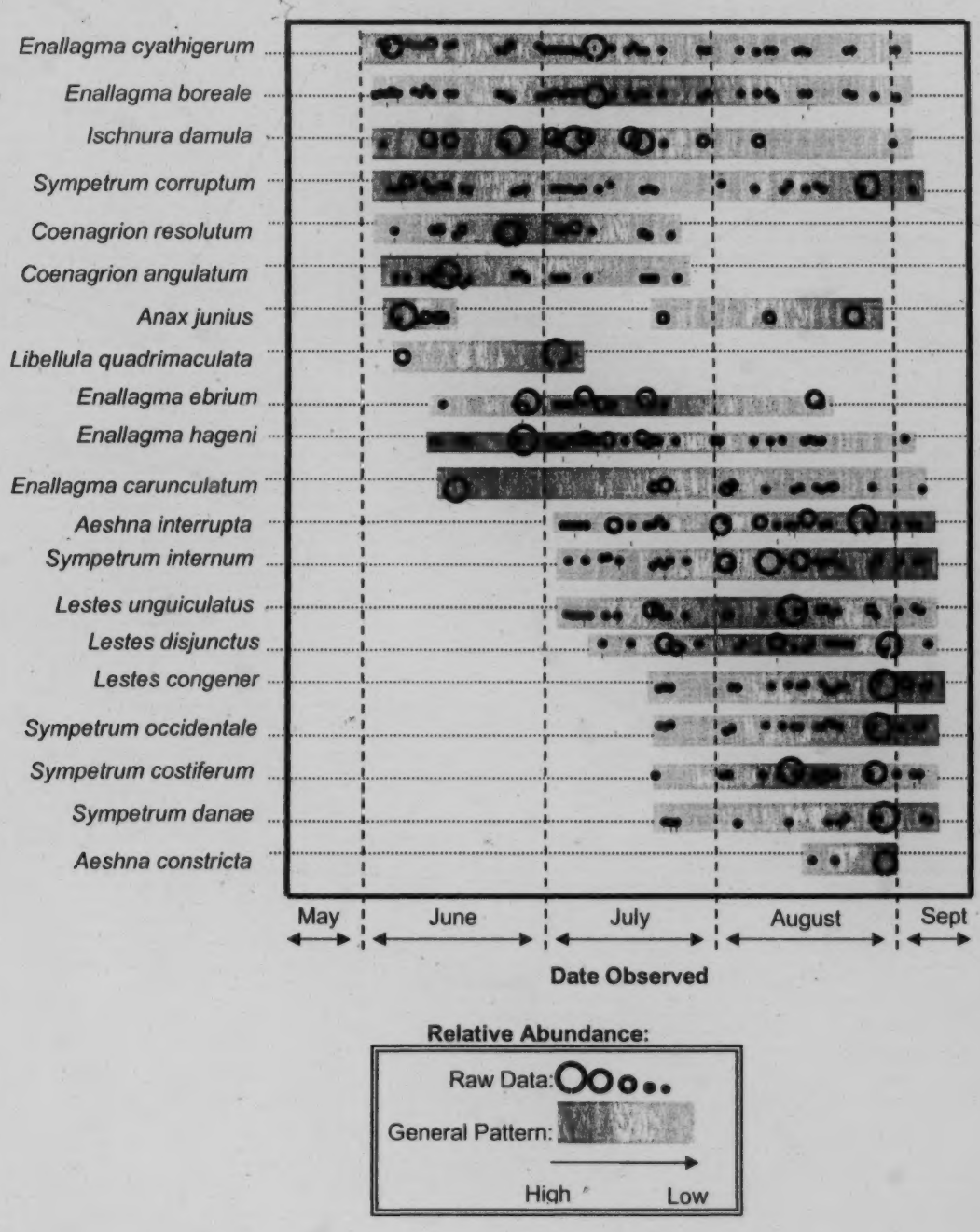
differences between treatments as well as to test for time effects between sampling periods. Where significant results were identified, I used Newman-Keuls *post hoc* multiple range test to isolate which means differed. Linear regression was used to investigate vegetation parameters affected by cattle grazing with the adult odonate community (i.e. abundance, richness, reproductive effort, and diversity). Non-normal and heteroscedastic dependent variables were log (y+1) transformed. Any $r^2>0.40$ were determined *a priori* to be robust, although results with $r^2<0.40$ thought to be biologically significant/interesting are also presented for discussion.

3.5 Results

3.5.1 Grazing impacts on wetland vegetation and their relevance to adult Odonates

Fifty-five different plants (Appendix-B) and twenty adult odonate species (with seasonal changes in composition) were detected within the study area (Figure 3-4).

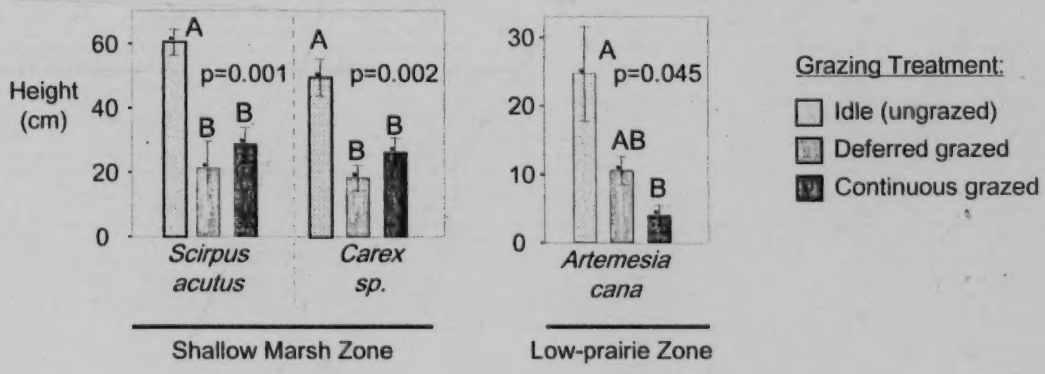
Figure 3-4: Seasonal changes in adult odonate composition and abundance, 2000.



3.5.1.1 Grazing and vegetation height

Plant height was significantly taller at idle compared to grazed wetlands for *Scirpus acutus* (Great Bulrush; F(2, 12)=14.77, p=0.001) and *Carex* spp. (Sedge; F(2, 14)=9.47, p=0.002) in the shallow marsh zone, as well as *Artemesia cana* (Silver Sagebrush (H(2)=6.20, p=0.045), in the low-prairie zone (Figure 3-5).

Figure 3-5: Impact of cattle grazing on wetland vegetation height.
(Bars represent ± 1 SE)



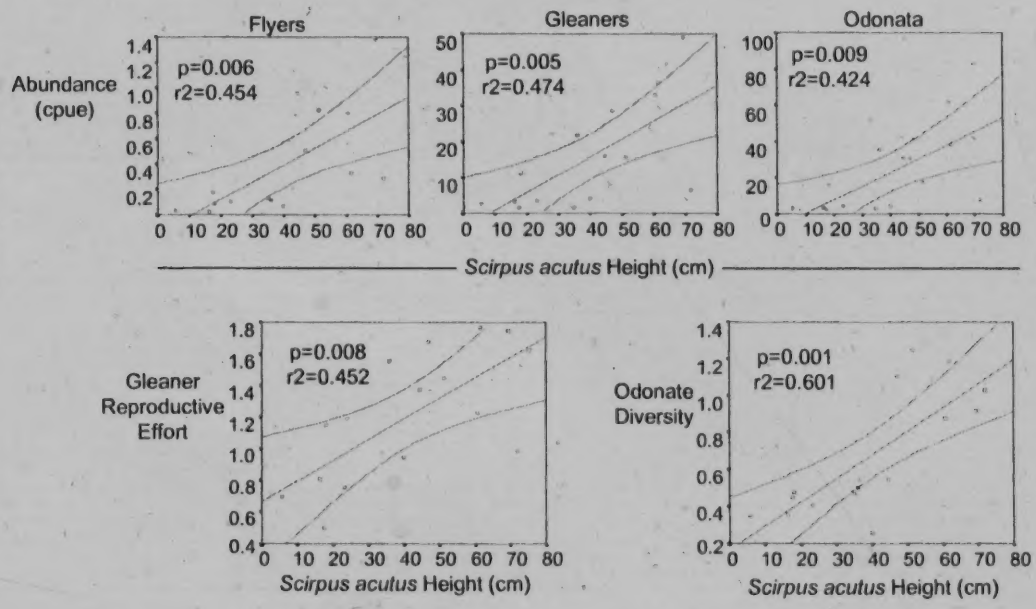
The height of dead vegetation (H(2)=7.34, p=0.025), *Agropyron smithii* (Western Wheatgrass; H(2)=10.98, p=0.004), and *Aster ericoides* (Tufted White Prairie Aster; F(2,11)= 21.98, p<0.001) in the low-prairie zone was also significantly higher at idle compared to grazed sites. No difference in height between treatments was detected for all other plants.

3.5.1.2 Vegetation height and odonates

The structure of *S. acutus* in the shallow marsh zone significantly impacted odonate abundance, reproductive effort, and diversity (Figure 3-6). This includes

higher Flyer (r^2=0.454, F(1,13)=10.82, p=0.006), Gleaner/Zygoptera (r^2=0.474, F(1,13)=11.72, p=0.005), and overall odonate abundance where taller stems occurred (r^2=0.424, F(1,13)=9.56, p=0.009). Gleaner reproductive efforts (r^2=0.462, F(1, 12)=9.91, p=0.008) and Odonata diversity (r^2=0.601, F(1, 13)=19.61, p=0.001) declined with decreased *S. acutus* height.

Figure 3-6: *Scirpus acutus* structure and the odonate community of a wetland. (Bars represent 95% Confidence Intervals)

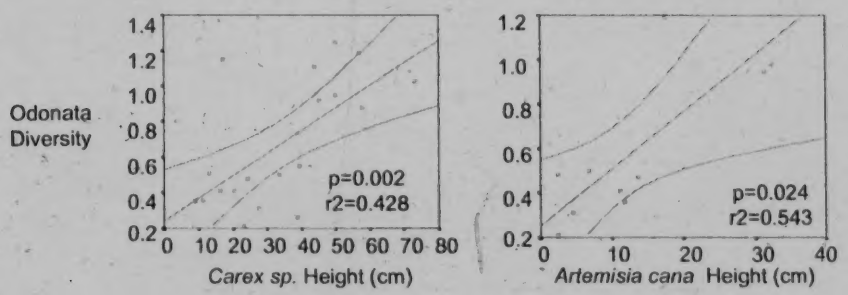


The height of *S. acutus* and *Carex* spp. in the shallow marsh, and *A. cana* in the low prairie zone directly affected four odonate species. Ae*shna interrupta* (Variable Darner) abundance declined with shorter *S. acutus* (r^2=0.455, F(1,13)=10.87, p=0.006) and *Carex* spp. (r^2=0.254, F(1,17)=5.78, p=0.028), and *Enallagma ebrium* (Marsh Bluet) abundance declined with shorter *Carex* spp.

(r^2=0.436, F(1,17)=13.13, p=0.002). *Aeshna constricta* (Lance-tipped Darner) and *Enallagma carunculatum* (Tule bluet) abundance declined with shorter *A. cana* (r^2=0.673, F(1,7)=14.4, p=0.007 for both).

Vegetation structure did not affect odonate richness, however odonate diversity was positively related to *Carex* spp. (r^2=0.428, F(1,17)=12.73, p=0.002) and *A. cana* height (r^2=0.543, F(1,7)=8.32, p=0.024) (Figure 3-7).

Figure 3-7: Odonate diversity and *Carex spp.* and *Artemisia cana* height. (Round 6 Shannon Diversity Index; Bars represent 95% Confidence Intervals)

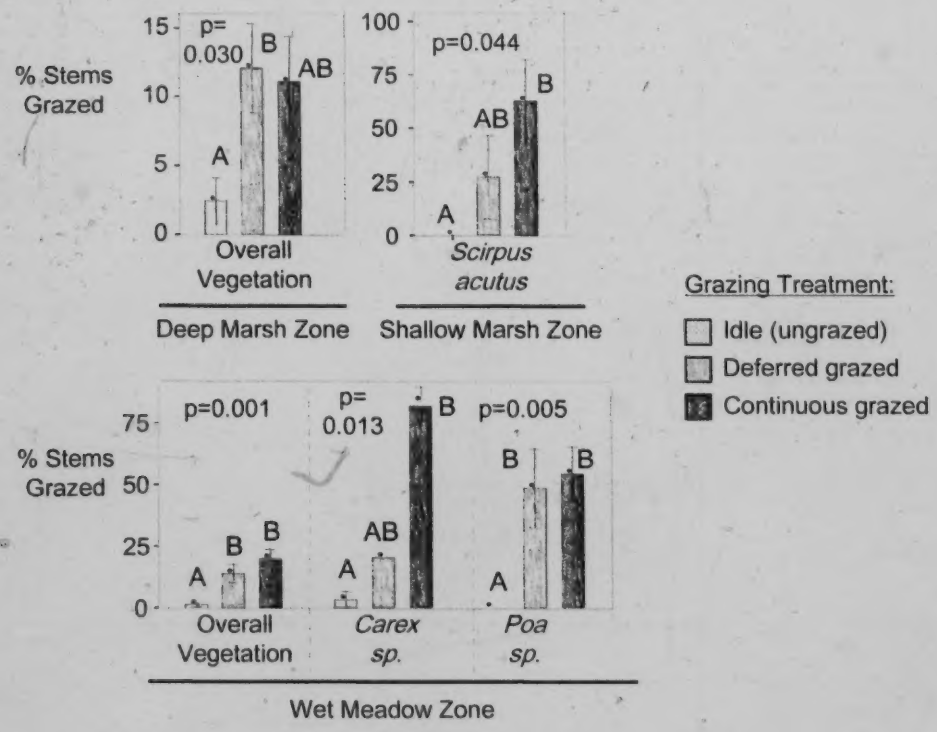


3.5.1.3 Grazing and percent stems grazed of wetland vegetation

The percent of stems grazed was significantly lower at idle wetlands (Figure 3-8). This includes overall vegetation (H(2)=7.02, p=0.030) in the deep marsh zone, *S. acutus* (H(2)=6.24, p=0.044) in the shallow marsh zone, overall vegetation (H(2)=15.00, p=0.001), *Carex* spp. (H(2)=8.61, p=0.013), and *Poa* spp. (Bluegrass; F(2,19)=7.15, p=0.005) in the wet meadow zone, as well as *A. smithii* (H(2)=15.13, p=0.001) and *Poa* spp. (H(2)=6.23, p=0.043) in the low-prairie zone. No difference

in percent stems grazed between treatments was detected for all other species of vegetation.

Figure 3-8: Percent stems grazed of wetland vegetation between treatments.
(Bars represent ± 1 SE; plots of untransformed data)



3.5.1.4 Percent stems grazed and odonates

No strong statistical relationship (i.e. r^2>0.40) existed between percent stems grazed and the abundance or reproductive effort of Odonata (with the exception of *E. ebrium*), however trends were consistent, biologically interesting, and noteworthy. The overall trend was a decrease in odonate abundance with increased percent stems grazed, including: *A. interrupta* and Flyers with *S. acutus* (shallow marsh zone); *E.*

ebrium with *Carex* spp. (wet meadow zone); and Flyers and Gleaners with overall vegetation in the wet meadow zone. There is a decline in abundance of *A. interrupta, E. ebrium,* as well as Flyers and Gleaners in general with increasing percent stems grazed of *Poa* spp. (wet meadow zone) and *A. smithii* (low-prairie zone) (see Table 3-2 for regression statistics).

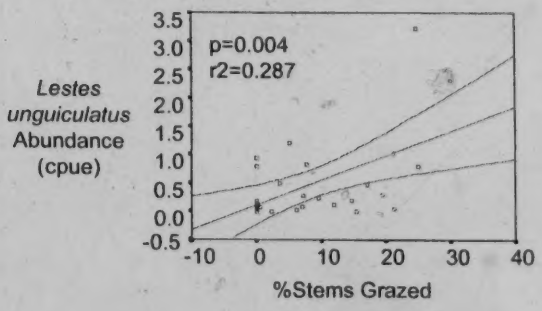
Odonate diversity was negatively related to percent stems grazed of overall vegetation (r^2=0.398, F(1,25)=16.51, p<0.001,) and *Carex* spp. (r^2=0.621, F(1,9)=14.74, p=0.004) in the wet meadow zone, as well as *A. smithii* (r^2=0.535, F(1,24)=27.56, p<0.001) and *Poa* spp. (r^2=0.525, F(1,23)=25.37, p<0.001) in the low-prairie zone.

Table 3-2: Simple linear regression results comparing wetland odonate fauna to grazing intensity (i.e. percent stems grazed) at a wetland.

Vegetation Zone	Independent variable (%stems grazed)	Dependent variable (odonate abundance)	r^2	F (df)	p-value
Shallow Marsh	*Scirpus acutus*	*Aeshna interrupta*	0.345	6.84 (1,13)	0.021
		Flyers	0.344	6.81 (1,13)	0.022
Wet Meadow	*Carex* spp.	*Enallagma ebrium*	0.397	5.96 (1,9)	0.038
	Overall vegetation	Flyers	0.183	5.58 (1,12)	0.026
		Gleaners	0.165	4.94 (1,25)	0.036
	Poa spp.	Flyers	0.290	8.17 (1,20)	0.010
		Gleaners	0.217	5.55 (1,20)	0.029
		Aeshna interrupta	0.312	9.08 (1,20)	0.007
		Enallagma ebrium	0.301	8.60 (1,20)	0.008
Low-prairie	*Agropyrnm smithii*	Flyers	0.180	5.26 (1,24)	0.031
		Gleaners	0.312	10.90 (1,24)	0.003

Lestes unguiculatus (Lyre-tipped Spreadwing) was the only odonate that showed a positive relationship to grazing intensity, increasing in abundance (r^2=0.287, F(1,25)=10.07, p=0.004) and reproductive effort (r^2=0.261, F(1,24)=8.47, p=0.008) with higher overall percent stems grazed in the deep marsh zone (Figure 3-9).

Figure 3-9: Positive relationship between the damselfly *Lestes unguiculatus* and the percent of overall stems grazed in the deep marsh zone.
(Bars represent 95% Confidence Intervals)



3.5.1.5 Grazing and percent cover of wetland vegetation

No significant difference in percent cover between grazing treatments was detected for any species of vegetation except the exotic *Circium arvense* (Canada-thistle; wet meadow zone), which had higher percent cover at ungrazed wetlands compared to deferred grazed wetlands (H(2)=9.06, p=0.011).

3.5.1.6 Vegetation percent cover and odonates

Overall odonate richness increased as the percent cover of *C. arvense* increased (i.e. highest at ungrazed compared to deferred grazed wetlands; r^2=0.423,

F(1,20)=14.68, p=0.001). No other significant relationship was found between *C. arvense* percent cover and odonate abundance, reproductive effort, or diversity.

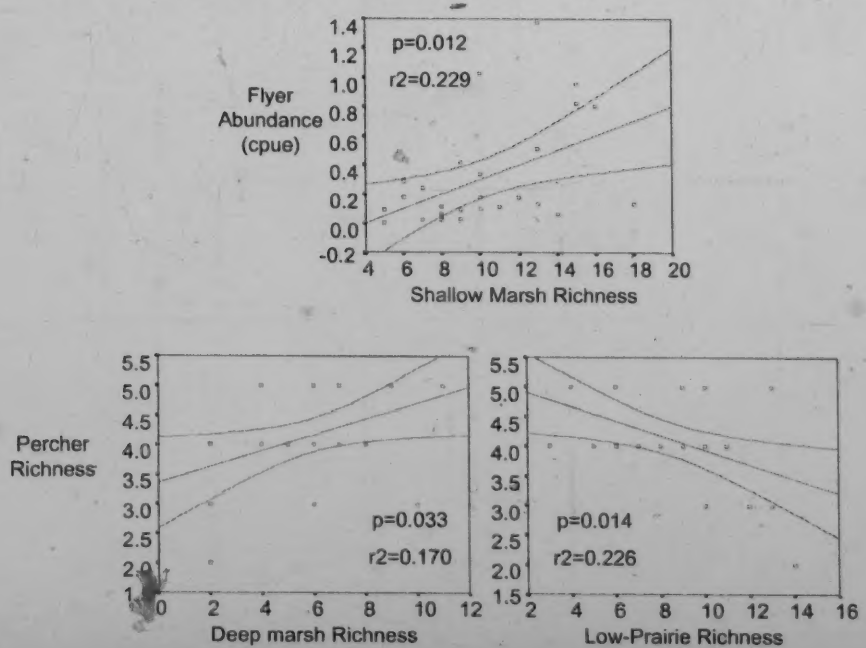
3.5.1.7 Grazing and wetland vegetation richness

No difference in vegetation richness was detected between grazing treatments, with a total of 24 – 29 species per treatment. No difference in exotic vegetation richness was detected between treatments.

3.5.1.8 Wetland vegetation richness and odonates

Vegetation richness was positively related to Flyer abundance in the shallow marsh zone (r^2=0.229, F(1,25)=7.41, p=0.012; Figure 3-10). Percher richness and vegetation richness were positively related in the deep marsh zone (r^2=0.170, F(1,25)=5.11, p=0.033) and negatively related in the low-prairie zone (r^2=0.226, F(1,24)=7.00, p=0.014; Figure 3-10).

Figure 3-10: Odonate abundance and vegetation richness at prairie wetlands.
(Bars represent 95% Confidence Intervals; plots of untransformed data)



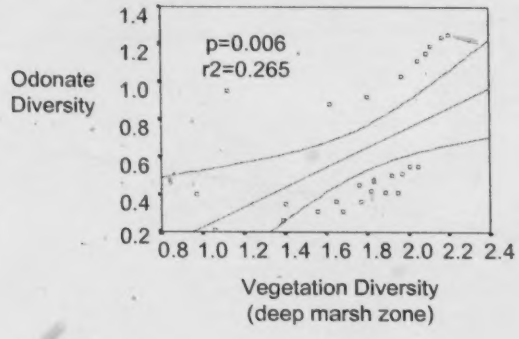
3.5.1.9 Grazing and wetland vegetation diversity

Vegetation diversity within each wetland zone was compared using Shannon diversity indices, which account for both the number and evenness of species in a community. No difference was detected (a=0.05).

3.5.1.10 Wetland vegetation diversity and odonates

Odonata and vegetation diversity in the deep marsh zone were positively related (r^2=0.265, F(1,25)=9.01, p=0.006; Figure 3-11).

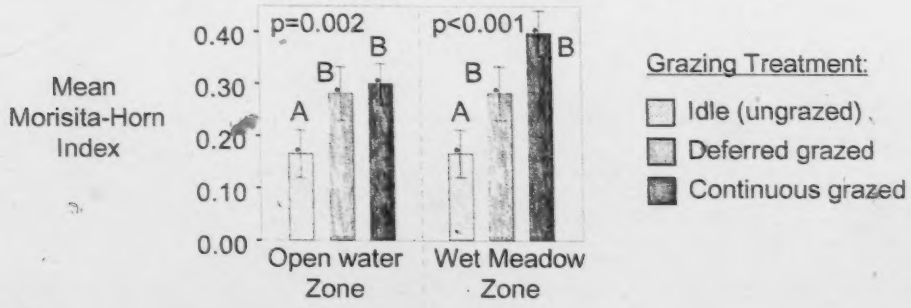
Figure 3-11: Odonate and wetland vegetation diversity within the deep marsh zone.
(Diversity = Shannon Diversity Index for Round 6; Bars represent 95% Confidence Intervals)



3.5.1.11 Grazing and wetland vegetation composition

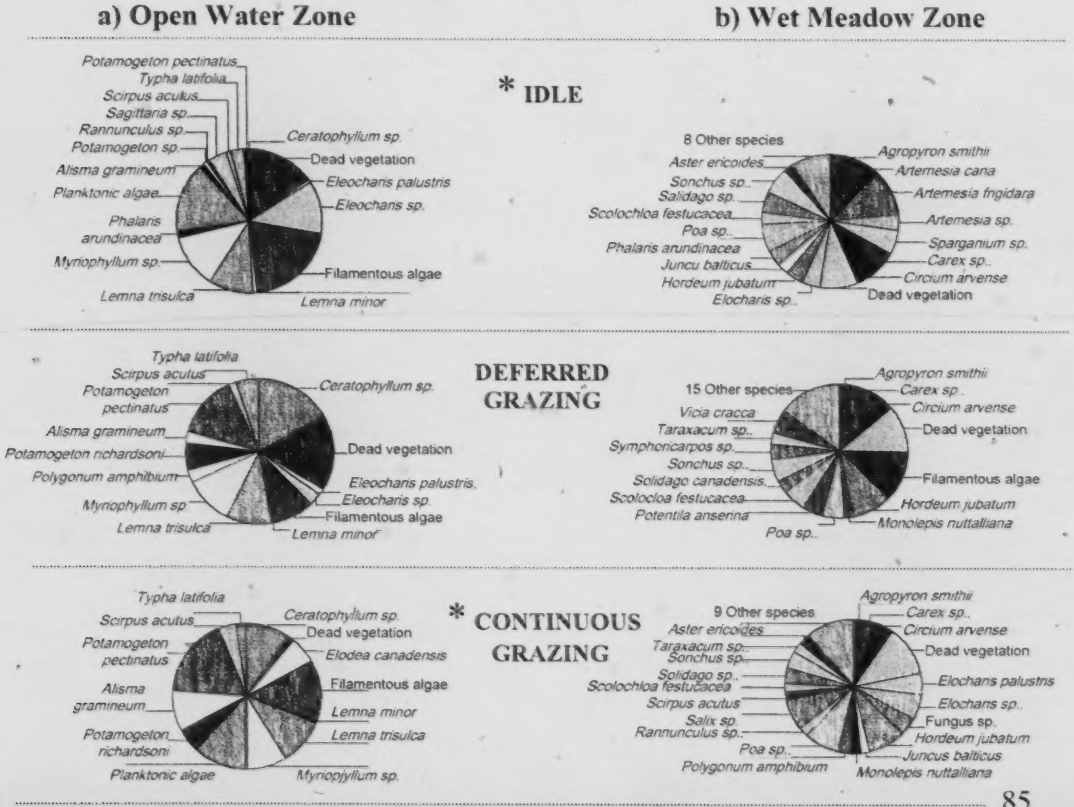
The similarity of vegetation composition between treatments was measured using the Morisita-Horn index, which is relatively independent of sample size or diversity (Wolda 1981). Significantly lower community similarity was detected at ungrazed compared to grazed sites, within both the open water and wet meadow zones, (H(2)=12.91, p=0.002 and H(2)=20.05, p<0.001 respectively; Figure 3-12).

Figure 3-12: Differences in vegetation composition among grazing regimes. (Bars represent ±1SE; plots of untransformed data)



This difference in composition corresponds to a reduction in dominant species at ungrazed control wetlands compared to disturbed grazed sites (Figure 3-13).

Figure 3-13: Vegetation composition and relative abundance between treatments. (Other= sum of all species <2% relative abundance; *= significant difference a=0.05)

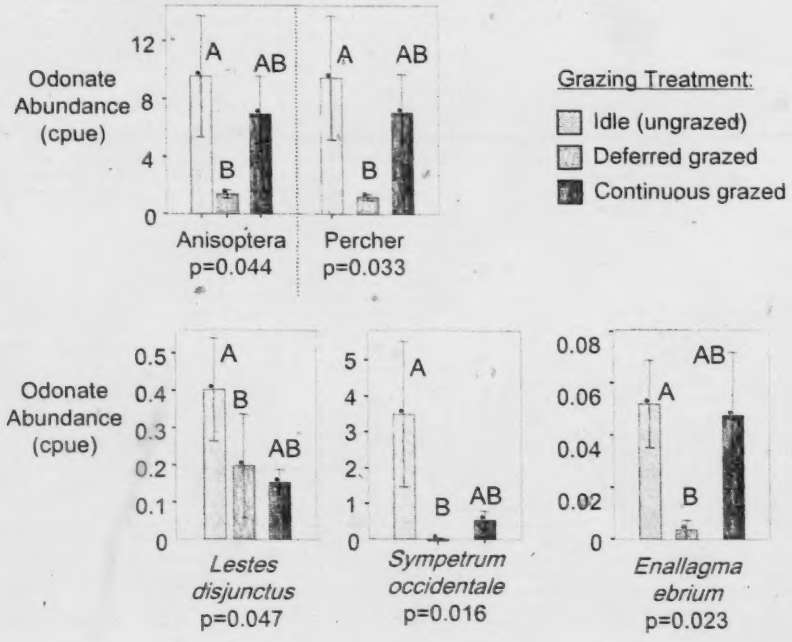


3.5.2 Grazing treatment and wetland odonate community

3.5.2.1 Grazing and odonate abundance

Significantly more Anisoptera (H(2)=6.23, p=0.044), Perchers (H(2)=6.84, p=0.033), *Sympetrum occidentale* (Western Meadowhawk, H(2)=8.31, p=0.016), *Lestes disjunctus* (Common Spreadwing, H(2)=6.12, p=0.047), and *E. ebrium* (H(2)=7.55, p=0.023) were detected at ungrazed compared to deferred grazed wetlands (Figure 3-14).

Figure 3-14: Odonate abundance between grazing treatments.
(Bars represent ±1 SE)

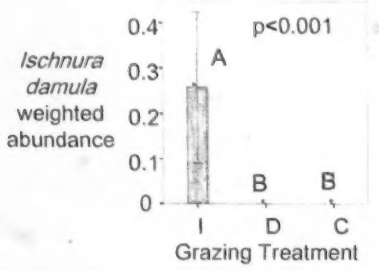


3.5.2.2 Grazing and odonate reproductive effort

The damselfly *Ischnura damula* (Plains Forktail, H(2)=6.47, p=0.039) was the only odonate that displayed significant differences in reproductive effort between

grazing treatments (Figure 3-15). The differences were quite marked with no reproductive behaviours observed at grazed sites, and reflect differences in observed abundance between treatments since almost all (i.e. 97%) *I. damula* records were from idle sites.

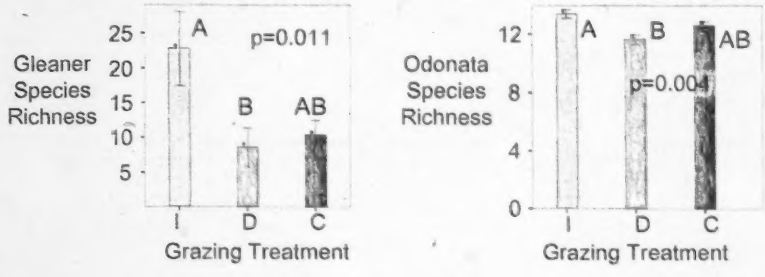
Figure 3-15: *Ischnura damula* reproductive effort between grazing treatments. (Bars represent ±1SE; I=idle, D=deferred, C=continuous; *= significant difference)



3.5.2.3 Odonate species richness

Species richness was significantly higher at ungrazed compared to grazed wetlands for both Gleaners and Odonata ($H(2)=9.05$, $p=0.011$ and $H(2)=10.86$, $p=0.004$ respectively, Figure 3-16.

Figure 3-16: Odonate richness between grazed and ungrazed wetlands. (Bars represent ±1 SE; I=idle, D=deferred, C=continuous)



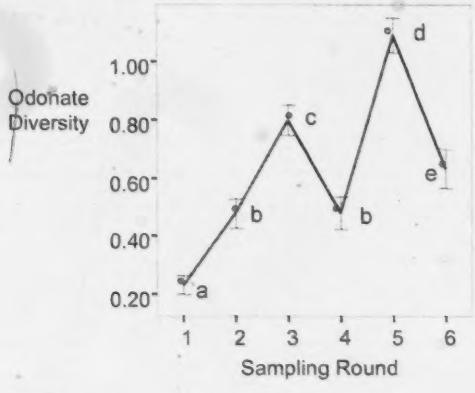
3.5.2.4 Grazing and odonate diversity

Odonate diversity was significantly different among sampling rounds but not significantly different between treatments when considering time (significant sampling period effects and interaction; no significant treatment effects; Table 3-3). Odonate diversity was significantly different for each round except Rounds 2 and 4, generally increased throughout the summer, and peaked during Rounds 3 and 5 (Figure 3-17).

Table 3-3: Results for differences in odonate diversity using Friedman's test. ($?^2$ values from Table B.1 in Zar 1999)

Source	df	H	$?^2_{crit}$	p-value
Treatment	2	2.092	5.991	not significant
Round	5	78.133	11.070	<0.05
Interaction	10	15.107	51.402	<0.05

Figure 3-17: Difference in adult odonate diversity between grazing treatments. (Shannon Diversity Index; Bars represent ±1 SE; a=0.05)



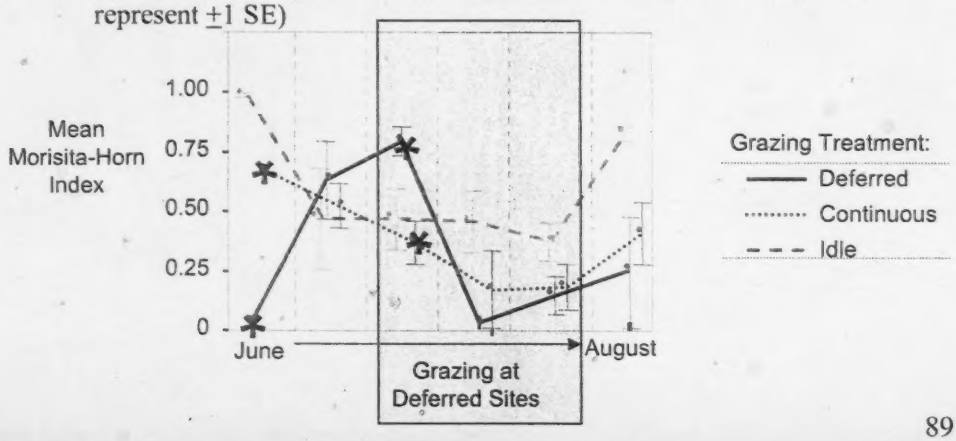
3.5.2.5 Grazing and odonate composition

Odonate composition was significantly different between grazing treatments when considering sampling period (significant treatment and sampling period effects, interaction not significant; Table 3-4). When cattle were present at grazed treatments (i.e. Rounds 1-5 for continuous sites; Rounds 3-5 for deferred sites) odonate community similarity was generally lower at grazed compared to ungrazed wetlands (Figure 3-18). Odonate community similarity was significantly higher at continuously grazed wetlands compared to deferred grazed wetlands during Round 1, however after one month of continuous grazing the reverse pattern was observed. Odonate community similarity was generally higher at ungrazed wetlands compared to grazed wetlands during the last three survey rounds.

Table 3-4: Results for differences in adult odonate similarity using Friedman's test. ($?_r^2$ values from table B.1 in Zar 1999)

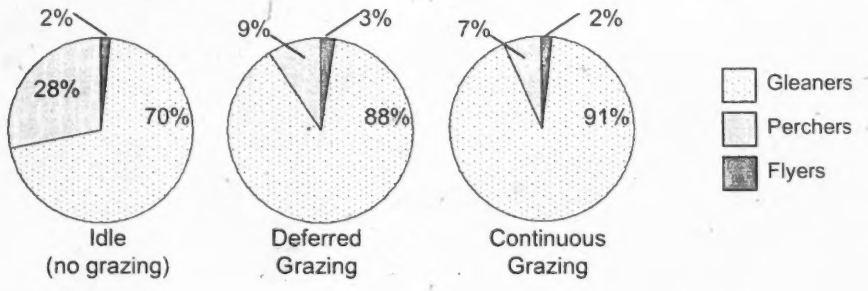
Source	df	H	$?^2_{crit}$	p-value
Treatment	2	7.276	5.991	<0.05
Round	5	17.083	11.070	<0.05
Interaction	10	15.107	18.307	not significant

Figure 3-18: Differences in adult odonate similarity between grazing treatments. (Morisita-Horn Index; *= significant difference when a=0.05; Bars represent ±1 SE)



Differences in odonate composition between treatments correspond to a general decrease in Perchers and increase in Gleaners at grazed wetlands compared to ungrazed wetlands (Figure 3-19).

Figure 3-19: Relative abundance of Perchers, Flyers, and Gleaners between grazing treatments.



3.6 Discussion

3.6.1 Odonate abundance and grazing

Both vegetation height (indirect grazing measurement) and percent stems grazed (direct grazing measurement) reflected the amount of grazing at sampled wetlands. The heights of a few key species of wetland vegetation were found to significantly affect the adult odonate community of a wetland, including *S. acutus*, *Carex* spp., and *A. cana* (Figures 3-5 and 3-6). Odonates were particularly negatively impacted when cattle grazed the emergent *S. acutus*. *S. acutus* grazing was associated with lower abundance for all odonates, particularly for Flyers and Gleaners. Adult odonates primarily use visual cues to assess overall breeding habitats, with limited use of tactile cues when selecting specific oviposition sites (Corbet 1999, Bernath et al. 2002). The significant change from the usually tall, lush *S. acutus* to shorter

grazed stems may be perceived by adult odonates as a decrease in habitat quality, and consequently fewer adults congregated, defended territories, and oviposited at that site. Additionally, the tall ungrazed *S. acutus* provided substantial refugia from the wind at the waters edge, and may explain my observations of significantly higher numbers of those odonates that are weak flyers (i.e. Zygoptera/Gleaners) at idle sites.

Flyers and most Gleaners are endophytic species (i.e. deposit eggs within vegetation; Needham et al. 2000, Westfall & May 1996) and both *A. interrupta* and *E. ebrium* were less abundant at sites with shorter *Carex* spp. stems in the shallow marsh zone (i.e. deferred grazed and continuously grazed wetlands, Figure 3-5). Additionally, Flyer abundance decreased with lower (shallow marsh) vegetation richness (Figure 3-10). This decrease in abundance may be due to an overall decrease in visual habitat cues, or these odonates may be specifically cueing in to the conditions of emergent sedges.

The height of vegetation in the low-prairie zone also affected the abundance of adult odonates at prairie wetlands. *A. constricta* and *E. carunculatum* were less abundant at sites with shorter *A. cana* stems (i.e. continuously grazed wetlands, Figure 3-5). Odonates congregate around water bodies during the day; however diurnal movement patterns dictate that nocturnal roosts, in addition to emergent vegetation, are necessary components of odonate habitat, especially for darners (Moore 1953). *A. cana* tends to grow in clumps and is one of the taller plants (i.e. 30-45 cm) in the low-prairie zone, and therefore may provide important roosting habitat for adult odonate in a landscape with limited roosting structures.

Oviposition is the link between terrestrial (adult) and aquatic (larval) odonates. Desirable oviposition sites can be predicted based on the habitat requirements of aquatic larvae since there is strong selective pressure for the female to select oviposition sites at which larvae survival is high (Buskirk & Sherman 1985). Other research has shown a positive correlation between adult odonate abundance and aquatic macrophyte cover (Clark & Samways 1996, Stewart & Samways 1998). No significant relationship was found between odonate abundance and percent cover for any species of wetland vegetation in this study. This may reflect true conditions, or insufficient data collection since percent cover estimates were conservative for deferred grazed sites (i.e. short-term intense grazing treatment). A two-week lag occurred between cessation of deferred grazing and vegetation surveys, thereby allowing for some level of plant re-growth and cover accumulation at these sites.

Physical disturbance from cattle, in addition to removal of visual cues resulting from grazing, may decrease the abundance of odonates in wetlands. Shoreline disturbances (i.e. waves from motorboats, intermittent dam releases) are thought to interrupt odonate emergence by removing, injuring, or killing emerging and very teneral odonates and making the shoreline itself a type of 'hostile' habitat (pers. comm. J. Acorn 2002). The physical disturbance by cattle as they walk through and browse emergent vegetation may disturb fragile emerging odonates and thereby decrease the abundance of odonates successfully emerging from a wetland.

3.6.2 Odonate reproductive efforts and cattle grazing

Gleaners (i.e. Zygoptera) reproductive effort was significantly higher at idle sites; these contained taller *S. acutus* stands (Figures 3-5 and 3-6). This is likely due to the oviposition or larval habitat requirements of Zygoptera. All damselflies rely on aquatic vegetation during oviposition since they are either epi- or endo-phytic (i.e. deposit eggs on or within vegetation; Corbet 1999). Additionally, all Zygoptera larvae are 'claspers' that crawl amongst submerged/emergent vegetation to avoid detection from predators and prey, and selective pressures are strong for female odonates to identify and select oviposition sites with high larvae survival rates (Buskirk & Sherman 1985).

I. damula was the only species that showed a significant difference in reproductive efforts between grazing treatments, with no reproductive behaviours observed at grazed sites despite the presence of both adult males and females. Members of the genus *Ischnura* are regarded to be especially weak flyers and individuals usually remain among the stems of the dense vegetation among the shoreline (Westfall and May 1996). Breeding *I. damula* may be cueing in to the shelter of the taller shoreline vegetation at ungrazed wetlands (i.e. *S. acutus* and *Carex* spp. in the shallow marsh zone, Figure 3-5).

Conversely, reproductive efforts and abundance for the damselfly *L. unguiculatus* were highest at wetlands with more intense grazing in the deep marsh zone (i.e. deferred grazed sites compared to idle sites, Figures 3-8 and 3-9). Lestes damselflies are adapted to ephemeral prairie wetlands (Westfall and May 1996) which are generally Class I-II with shorter, less dense vegetation compared to the

more permanent Class IV-V wetlands in this study. *L. unguiculatus* may be cueing in to the less dense emergent vegetation at deferred grazed sites, which more closely resembles typical *L. unguiculatus* habitat (i.e. open ephemeral ponds) than the dense, tall emergent vegetation of ungrazed wetlands (Westfall and May 1996).

3.6.3 Odonate species richness and grazing

Total Odonata and Gleaner species richness was significantly higher at ungrazed wetlands compared to deferred grazed wetlands (i.e. intense short-term grazing treatment; Figure 3-10). Other studies have also shown a negative correlation between adult odonate species richness and grazing intensity (as measured by percent stems grazed; Hornung & Rice 2003) or shoreline trampling from grazing (by African buffalo; Stewart & Samways 1993).

Percher richness is positively related to emergent vegetation richness in prairie wetlands (Figure 3-10). The importance of emergent vegetation to higher odonate species richness is consistent with research in other regions where adult Odonata richness and reed cover were positively correlated (Stewart & Samways 1998). Percher richness decreases as vegetation richness in the low-prairie zone increases. The significance of low-prairie species richness to Perchers may indirectly reflect grazing intensity since upland vegetation richness is known to increase with grazing intensity (Collins 1984). Additionally, *Sympetrum* spp. (i.e. Perchers) were observed spending a larger portion of the day in upland habitat compared to other odonates (i.e. congregating at water edge later in the day compared to other odonates), and may therefore be more affected by changes to the low-prairie zone than other odonates.

C. arvense was the only plant that showed a significant difference in percent cover between grazing treatments. The positive relationship between *C. arvense* percent cover and Odonata richness may be spurious, or may be a very indirect measure of cattle grazing. *C. arvense* was more abundant (i.e. percent cover) at idle compared to deferred grazed sites within the study area. These were unexpected results since *C. arvense* is considered an unpalatable invasive forb that is expected to increase due to overgrazing (Alberta Agriculture, Food, and Rural Development 1996), and restricting grazing to allow interspecific competition is a recommended management practice for controlling C. arvense (Edwards et al. 2000).

3.6.4 Odonate diversity and grazing

Odonata diversity was affected by the height of two vegetation species; odonate diversity increased with taller *S. acutus* and *A. cana* stems (Figure 3-6 and 3-7). The height of both *S. acutus* and *A. cana* affected the abundance of four odonate species, including *A. interrupta*, *A. constricta*, *E. ebrium*, and *E. carunculatum* (i.e. positive relationship between stem height and odonate abundance). Lower odonate diversity at wetlands with shorter *S. acutus* and *A. cana* stems (i.e. grazed wetlands) may reflect the decline or absence of the above odonate species due to a lack of suitable habitat cues that these two vegetation species may provide, such as ovipositing substrates or wind-shelter (i.e. *S. acutus*), and nocturnal roosts (i.e. *A. cana*).

Odonata diversity declined as grazing intensity increased on overall wet meadow vegetation, *Carex* spp., *Poa* spp., and *Agropyron smithii* (i.e. percent stems

grazed). Grazing is uneven within a pasture and cattle with unrestricted access to wetlands tend to focus their grazing efforts on riparian vegetation and other preferred forage species (Fitch & Adams 1998). *Carex* spp., *Poa* spp., and *A. smithii* are considered palatable good quality forage for cattle and may therefore be indicative of cattle utilization of a pasture (Alberta Agriculture, Food, and Rural Development 1996).

Adult odonate diversity is positively related to emergent vegetation diversity (i.e. deep marsh zone). Adult odonates require aquatic vegetation for emergence substrates, perch platforms, and oviposition cues, all of which are activities that occur at the water's edge (i.e. deep marsh zone). More diverse vegetation templates may offer a greater variety of vegetation habitat and therefore accommodate different odonate species with different microhabitat requirements.

Odonate diversity varies significantly (i.e. two distinct pulses) over the summer season; first as early emerging species emerge and mature and the second, for late-emerging species (Figure 3-17). Adult odonate diversity was not significantly different between grazing treatments (Table 3-4). My results do not support the Intermediate Disturbance Hypothesis (IDH) wherein maximum species diversity occurs in between the states of extreme disturbance (Connell 1978). Extreme states in my study were chronic disturbance at continuously grazed sites and greater stability (i.e. no grazing disturbance) at idle wetlands. Moderate levels of cattle grazing have been shown to increase vegetation diversity since unpalatable (ungrazed) forbs flourished because they were released from competition from dominant grasses (Hickman & Hartnett 2002). Results in support of the IDH were

found for larval odonates (this study) as well as adult odonates across a gradient of disturbance in lotic habitats (Stewart & Samways 1998). Although diversity indices are similar between treatments, it is important to look at differences in odonate species composition since it is possible that overall diversity remains unchanged but a shift in composition has occurred.

3.6.5 Adult odonate composition and grazing

Adult odonate composition significantly changes over the summer as the flight season for early emerging species ends and late emerging species begins (Figure 3-4, Table 3-4). Odonate composition is more similar within ungrazed sites compared to within deferred grazed or continuously grazed sites, especially during late summer (Figure 3-18). The similarity of odonate fauna within deferred grazed sites is high at the on-set of deferred grazing, but quickly drops and remains low after cattle are present. The odonate community at ungrazed wetlands consists of relatively more Perchers compared to deferred grazed or continuously grazed wetlands, which suggests a shift in odonate community composition with the presence of grazing disturbance (Figure 3-19).

3.7. Summary and recommendations

Some of the known 'ecological costs' of cattle grazing are decreases in the diversity and abundance of certain taxa, changes in community organization, changes in the physical characteristics of terrestrial and aquatic ecosystems, and disruption of ecosystem function (i.e. nutrient cycling, succession) (Fleischner 1994). Although

this study did not attempt to address the impact of grazing on ecological function, grazing was shown to change wetland vegetation composition and structure, which in turn decreased odonate abundance and diversity, and altered odonate composition. Cattle grazing significantly impacted emergent vegetation structure, but did not influence vegetation richness or diversity, therefore my results support the hypothesis that cattle grazing impacts wetland vegetation structure (i.e. accept Hypothesis #3).

Results from this study show that adult odonates respond to changes in vegetation structure due to unrestricted cattle grazing. Cattle-altered vegetation subsequently decreased odonate abundance, thereby supporting the hypothesis that wetland vegetation impacts wetland adult odonate abundance (i.e. accept Hypothesis #4). The height and diversity of emergent vegetation, especially *S. acutus*, had the greatest impact on odonate abundance and reproductive effort, especially for Gleaners (Suborder Zygoptera) which are epiphytic ovipositors, weaker flying odonates that tend to occupy the reeds of emergent vegetation. Overall odonate diversity was affected by the height of three key species of vegetation (i.e. *S. acutus, Carex* spp., and *A. cana*) highlighting the importance of the structure of both emergent vegetation (i.e. breeding habitat) and upland vegetation (i.e. nocturnal roosts or foraging sites) to the life history of prairie odonates. Many of the results suggest that non-linear regressions may better explain patterns in the data than simple linear regression. Future studies should consider non-linear regression analysis, as well as measure aquatic macrophytes in more detail since submergent vegetation provides critical habitat for both larval (i.e. cover) and adult (i.e. oviposition substrate) odonates.

Future research should also incorporate exuvia collections in order to get a more accurate indication of odonate breeding success at a wetland.

Results also suggest that wetland vegetation structure is more important than vegetation composition to the life history of odonates. Odonates did not respond to changes in wetland vegetation composition due to grazing, yet odonate abundance, reproductive effort, and diversity were all significantly affected by changes in vegetation height due to cattle grazing. Vegetation diversity positively impacts odonate diversity; however vegetation diversity is more a consequence of wetland class rather than grazing treatment.

Although odonates responded to quantitative measurements of cattle-altered vegetation, few significant results were found when the wetland odonate community was directly compared to grazing treatment (i.e. qualitative variable). This counter-intuitive result may be due to simplistic characterization of grazing treatments (grazing treatment was a categorical variable assigned based on the duration of cattle in a pasture, i.e. all summer, part of summer, or altogether absent). Future studies should be more sensitive to the complexities of grazing regimes (i.e. number of cattle in pasture, size of pasture, number of wetlands in pasture). If it is logistically impossible to control/alter existing grazing regimes to attain more precisely defined grazing treatments (as in this study), then future studies should quantify grazing treatments as a continuous variable (i.e. AUM's, # cattle/ha pasture, or percent utilization).

3.8 Literature cited

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Chapter 4

Odonates as biological indicators of the aquatic macro-invertebrate community and overall habitat quality at grazed prairie wetlands

4.1 Introduction

Biological Indicator Theory has been extensively studied in the context of lotic habitats and aquatic macro-invertebrates are widely accepted and implemented as suitable habitat indicators in streams and rivers (Resh et al. 1995, Reynoldson et al. 1997, Karr & Chu 1997, US EPA 2002a & b), but little is known as to which, if any, taxa are most sensitive and suitable as biological indicators in wetlands.

Prairie wetlands are typically high-nutrient, high-productivity systems adapted to recurring natural drought cycles every 5-20 years (Van der Valk & Davis 1978). The wetlands investigated in this study are not typical ephemeral prairie wetlands since water levels are maintained for irrigation and cattle watering sites all summer. Cattle are allowed unrestricted access to wetlands and target their grazing efforts on highly palatable emergent vegetation (Fitch & Adams 1998).

Rotational grazing regimes, in which hundreds of cattle are moved through a series of pastures throughout the summer season, are common in the study area; however, smaller cattle operations exist where relatively fewer cattle (i.e. <100 cattle) graze one pasture and wetland all season. Cattle grazing primarily affects vegetation (i.e. richness, abundance, composition, and structure), but may also alter soil structure or ecosystem processes such as nutrient cycling (Fleischner 1994).

4.2 Objective

I investigate the impact of three cattle grazing regimes on the aquatic macro-invertebrate communities of prairie wetlands, and integrate biotic and abiotic data to assess the suitability of odonates (Insecta: Odonata) as a biological indicator of grazing disturbance at prairie wetlands.

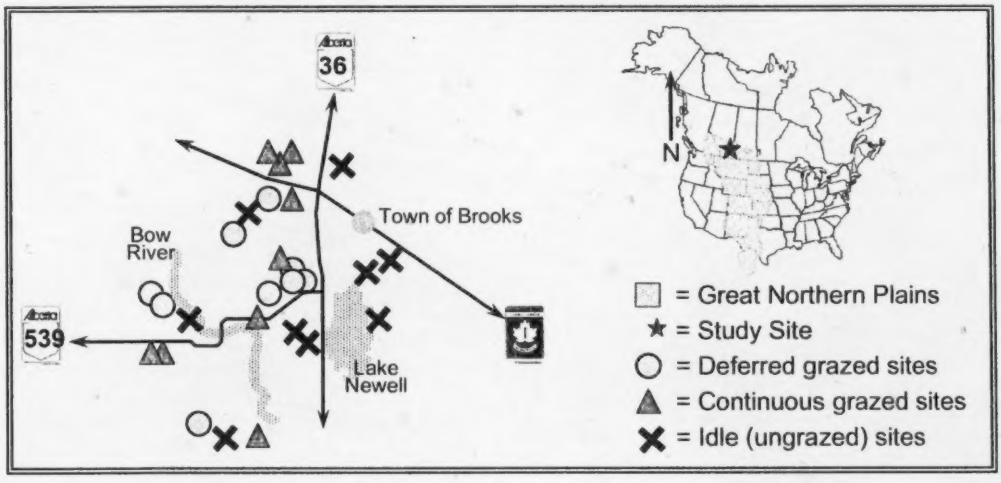
4.3 Study area

Study sites are located within an 1800 km^2 area of the dry mixed-grass prairie in south-eastern Alberta, Canada (50° 30' N, 111° 55' W) (Figure 4-1). Agriculture (i.e. cropland, irrigation infrastructure, beef cattle grazing) and oil and gas extraction are the main contemporary sources of income generation and concomitant land disturbance within the study area. Free-roaming bison herds and wildfires were past sources of landscape disturbance, but these have been eliminated as 94% of Alberta's native grasslands have been converted to cropland (Environment Canada 2002). The dry mixed-grass prairie still supports substantial tracts of grazing land because precipitation is insufficient for dependable cropping regimes.

Moisture deficits are typical in this dry and windy region, particularly at the end of summer (i.e. 250mm mean annual precipitation; 19 km/h mean annual wind speed) (Environment Canada 2002). Prairie wetlands are highly variable and many are dry by the end of summer. Inter-annual variation is significant and follows a 5-20 year drought cycle (van der Valk & Davis 1978). However, the sites in this study are intensively managed and are not natural ephemeral wetlands. The Eastern Irrigation District (EID) and Ducks Unlimited Canada (DUC) intensively manage wetlands to

serve as irrigation basins, sources of drinking water for beef cattle, and to a lesser degree wildlife habitat, primarily for waterfowl or shorebirds. Water levels are artificially controlled and wetlands are kept unnaturally high and stable (i.e. wet all season). Decreased hydrologic variation and deep flooding results in a subsequent increase in vegetation diversity (Stewart and Kantrud 1971), an increase in the concentration of nitrogen and phosphorus (Kadlec et al. 2000), and a shift in the typical aquatic invertebrate fauna (Murkin and Ross 2000).

Figure 4-1: Study area location and distribution of study sites.



4.4 Methods

4.4.1 Experimental design

Wetlands were selected based on five criteria: grazing regimes during the previous five years, basin characteristics (i.e. size, source, and degree of isolation represented at least once in each treatment), surrounding habitat (i.e. grasslands only,

no croplands), wetland class (i.e. Stewart and Kantrud (1971) Class IV or V), and access permission.

Manipulating pasture sizes and stocking rates for the sake of this project was logistically infeasible. Continuous and rotational grazing are the two predominant grazing strategies employed within the study area. Continuous grazing regimes keep cattle in one pasture all season (i.e. May to August) and consequently involve small herds of cattle on small sections of private land. More commonly, rotational grazing merges many herds to form a single 'super herd' managed by a local grazing association. To ensure pasture re-growth and a constant supply of forage, cattle are directed through a series of pastures beginning with early-germinating tame pastures (i.e. exotic species) and ending on native grasslands. Deferred grazing regimes are a derivation of rotational grazing systems and are promoted by DUC in an attempt to decrease waterfowl nest exposure and mortality due to foliage removal and cattle trampling. DUC negotiates contracts with grazing managers to delay cattle access to specific wetlands until after July 15th, at which time most waterfowl broods have fledged (NAWMP 1999).

The experimental units for this study were the individual wetlands. Wetland characteristics were estimated from a continuous 250m stretch of shoreline that was selected to best represent the variation in wetland vegetation and shoreline structure. All data collection took place along these 250m segments (delineated into six 50m sections selected as random sub-samples) to standardize sampling wetlands of various sizes. The grazing regime in the pasture surrounding each wetland supported one of three treatments: deferred grazing (i.e. mid-July to mid-August), continuous grazing

(i.e. approximately May to August), and idle (ungrazed) pastures (i.e. control treatment). There were six sampling rounds in 2000 and three sampling rounds in 2001, each at 2-week intervals. Stratified random design was used for all data collection (Krebs 1989): strata were delineated according to wetland vegetation zones according to Stewart and Kantrud (1971) for vegetation and adult odonate surveys, and distance from vegetation/water interface for all aquatic macro-invertebrate and water collections. Pseudoreplication was avoided since experimental units (i.e. wetlands) were replicated nine times in each of the three treatments, and each wetland was far enough apart (i.e.>1km) to be considered statistically independent relative to dispersal distances (Hurlbert 1984). Grazing was ubiquitous throughout the study area and as a result, the limitation on sample size was the number of ungrazed (idled) pastures in local protected areas. A total of twenty-seven wetlands were selected for a balanced design of nine replicates in each of the three treatments (Figure 4-1). Response variables included wetland odonate (adult and larvae) and aquatic macro-invertebrate community metrics, vegetation structure, and nitrogen and phosphorus concentrations.

4.4.2 Measuring the vegetation community

Vegetation surveys were conducted once per site at the end of summer in 2000 when the majority of plants had matured and were more easily identified. Grazing estimates were conservative for deferred sites because surveys occurred after grazing ceased, allowing for about two weeks of re-growth, whereas, at continuously grazed sites grazing was usually ongoing during surveys.

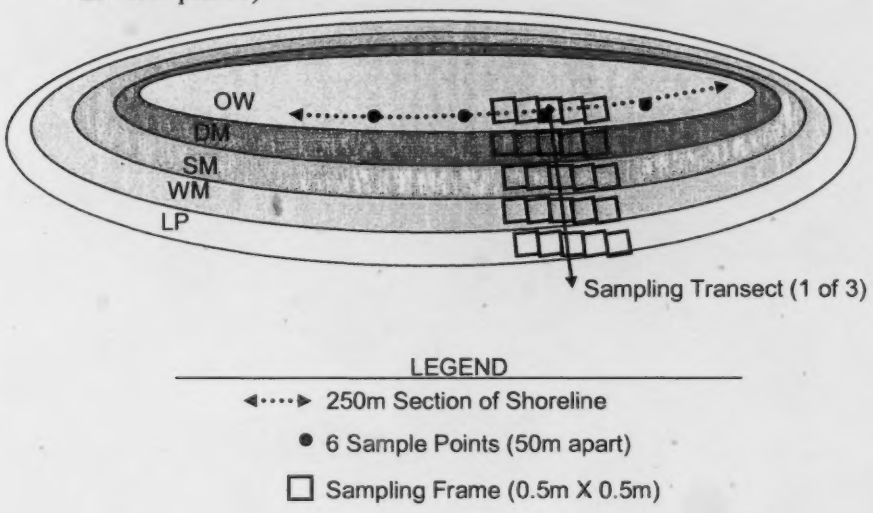
I measured vegetation characteristics relevant to the life histories of odonates including structure (i.e. height, abundance) and composition. Percent cover was estimated as a measure of plant abundance since it is the most common metric for estimating the percent of ground surface covered by the projection of a plant (NARSC 1996). I used a Robel pole to measure plant height since it is the simplest and most accurate method for integrating both vegetation structure and density in prairie habitats (NARSC 1996, Ganguli et al. 2000). Grazing intensity is most often measured as percent utilization of palatable forage species, but I chose percent stems grazed for each plant species as a quantitative index of grazing intensity since I was interested in the impact of cattle grazing on the entire vegetation community rather than select palatable species (Holechek et al. 2001). I used sampling frames to measure percent cover, percent grazed stems, and vegetation composition (NARSC 1996), and chose a $0.25m^2$ quadrat (i.e. 0.5m by 0.5m) for this study since survey lines included both tall/dense and short/sparse vegetation (USEPA 1997, Daubenmire 1959). Most plants were identified to species except for willows and some non-flowering forbs (see Appendix-B for complete list).

The wetland vegetation community was composed of different zones of vegetation in concentric rings outward from the water's edge, influenced by the degree of water permanency (Stewart & Kantrud 1971). Due to the inherent decrease in vegetation height and abundance as distance from water increased (i.e. tall vigorous emergent vs. short sparse upland plants), comparisons between treatments were made within each of the five vegetation zones defined by Stewart and Kantrud

(1971), including: open water, deep marsh, shallow marsh, wet meadow, and low-prairie (Figure 4-2).

Three of the six permanent survey points at each wetland were randomly chosen as starting points for vegetation transects. Rope transects were laid out perpendicular to the shoreline encompassing a span from 2m inside the open water zone outward to the beginning of the upland prairie zone, thereby traversing all wetland communities (Figure 4-2). Five sub-samples of quadrat and Robel pole measurements were taken from the middle of each vegetation zone, resulting in about 75 sub-samples per wetland.

Figure 4-2: Vegetation survey design.
(OW=open water, DM=deep marsh, SM=shallow marsh, WM=wet meadow, LP=low-prairie)



4.4.3 Measuring water quality

Budget constraints dictated that the minimum number of water samples be submitted for analysis. All samples were collected three or more days after a storm

event to minimize effects from run-off. Three of the six survey points were randomly selected (Figure 4-2), and 1L of water was collected from each point at 1m into the open water zone at a depth of 30cm. The three samples were combined and one 0.5L composite sample was packed in ice and shipped that day to the Limnology Lab at the University of Alberta.

Two water samples were collected from all wetlands: 1) before deferred grazing began (i.e. mid-July 2000), herein called pre-cattle sample, and 2) during deferred grazing (i.e. late-August 2000), herein called post-cattle sample. These samples were analysed for ammonium (NH_4+), nitrates and nitrites (NO_2+NO_3), total Kjeldahl nitrogen (TKN), total phosphorus (TP), total dissolved phosphorus (TDP), total dissolved solids (TDS), and Chlorophyll-a concentrations (i.e. algae productivity).

4.4.4 Collecting larval odonates and other aquatic macro-invertebrates

During each sampling round, odonate larvae were collected along with other aquatic macro-invertebrates using a D-frame sweep net (one of the best devices for sampling aquatic invertebrates in heavily vegetated wetlands; Turner & Trexler 1997). Three of the six survey points were randomly selected, and three distinct micro-habitats were sampled at each point including: emergent vegetation (i.e. 1m outward from open water), emergent vegetation /open water interface, and deeper open water (i.e. 1m inward from vegetation/water interface). Five 1m sweeps were collected from each microhabitat for a total of forty-five sweeps per wetland. Aquatic invertebrates that were captured were preserved in 70% ethanol solution and were

identified later in the laboratory at the University of Alberta. Samples from the year 2000 were identified to Family with the exception of subclass Acari (identified to subclass only), and orders Odonata and Ephemoptera (both identified to genus), whereas 2001 invertebrates were identified to the lowest identifiable taxonomic level and used for all richness and diversity calculations (i.e. Mollusca and Annelida were identified to genus, Class Crustacea were identified to species, Class Arachnida were identified to subclass, and Class Insecta were identified to family (i.e. Orders Diptera and Hemiptera) or genus (i.e. Orders Coleoptera, Ephemoptera, and Trichoptera); see Appendix-C for complete taxa list). Primary references for identification were Clifford (1991) and Merritt and Cummins (1996).

4.4.5 Surveying adult odonates

Wetlands were surveyed at two-week intervals during the summer of 2000 (six sampling periods) and 2001 (three sampling rounds) to account for seasonal changes in odonate fauna. Odonates display diurnal movement patterns by roosting in upland vegetation at night and concentrating around water bodies during the day. Diurnal patterns may be predicted at an even finer scale since mature males concentrate around the water from morning to afternoon, females make shorter trips to and from the water throughout the day, and immature males generally avoid riparian habitat until late in the afternoon when the density of mature males decreases (Moore 1953). Survey biases from the diurnal movement of odonates were avoided by randomizing the time of day at which surveys were conducted at each site and each sampling period.

Standardized survey methodology for adult odonates involved direct counts made while walking through riparian habitat during suitable flight conditions (i.e. low winds, warm, sunny). I used a hand-net to catch, identify, and release odonates (Moore 1953). Disturbance by the surveyor caused a flight-response making any sedentary odonates visible and thereby facilitating identification (Moore 1953). Each wetland vegetation zone was searched once in a serpentine-transect fashion. Abundance estimates were standardized based on time spent searching (i.e. initially 1½ hours per site, reduced to 45 minutes in 2000) and catch-per-unit-effort (CPUE) was calculated as the number of individuals counted/time spent searching.

Direct counts are estimates of the number of odonates at a 250m section of wetland. Despite a tremendous reservoir of individual odonates surrounding riparian habitats, odonate density in riparian/breeding habitat has been shown to remain fairly consistent due to territorial behaviour (Moore 1953). Direct counts are considered a conservative measurement of odonate abundance (Conrad et al. 1999).

4.4.6 Statistical methods

Prior to analysis, normal distributions were tested using the Shapiro-Wilk test, and homogeneity of variances were tested for using the Levene statistic (SPSS 1999, Zar 1999). An *a priori* alpha of 0.05 was selected for all statistical tests. Aquatic macro-invertebrate collections were more frequent in 2000 (i.e. six rounds vs. three rounds in 2001), and more precisely identified in 2001 (i.e. lowest identifiable taxonomic level vs. family level identification in 2000); therefore all analysis regarding invertebrate abundance used 2000 data, and all analyses concerning

richness or diversity measures used 2001 data. Friedman's test (nonparametric repeated measures) was used to test for differences in wetland aquatic macro-invertebrate community between grazing treatments. Where significant results were identified, I used Newman-Keuls *post hoc* multiple range test to isolate which means differed. Simple linear correlation analysis was used to investigate the relationship between larval odonates and other aquatic macro-invertebrates in preference to regression analysis because there is no clear dependent variable when considering these two communities; Spearman's rank correlation was used since data were non-normal. I used SPSS version 11.0 (SPSS 1999) for all analyses except Friedman's test, which was performed using SYSTAT Version 10 (SYSTAT 2000).

Direct ordination analysis was used to describe patterns in wetland odonate community in relation to environmental data, including water quality, vegetation structure/community, and aquatic macro-invertebrate community metrics. Adult and larval odonate data were analyzed separately using CANOCO 4.0 (1997 version). Spearman's rank correlation analysis was used *a priori* to identify highly correlated parameters (i.e. >0.70) within both species and environmental data matrices to guard against these variables biasing the ordination results. All species and environmental data were standardized via relativization by maxima, and non-normal data were log (x+1) transformed (McCune & Grace 2002). Automatic forward selection of environmental variables was used together with a Monte Carlo permutation test (full model) to identify those parameters that significantly contribute to the ordination, to reduce the large environmental data set (i.e. only significant environmental parameters were included in the final ordination analysis), and consequently

restricting these ordinations to exploratory rather than hypothesis-based analysis (McCune and Grace 2002). Redundancy Analysis (RDA) was selected since it is an extension of multiple linear regression (Makarenkov & Legendre 2002). The statistical significance for the relationship between species data and the first ordination axis, as well as species data and the set of environmental variables (i.e. sum of all eigenvalues) was computed using Monte Carlo permutation test (i.e. full model to minimize type II error and maximize statistical power).

4.5 Results

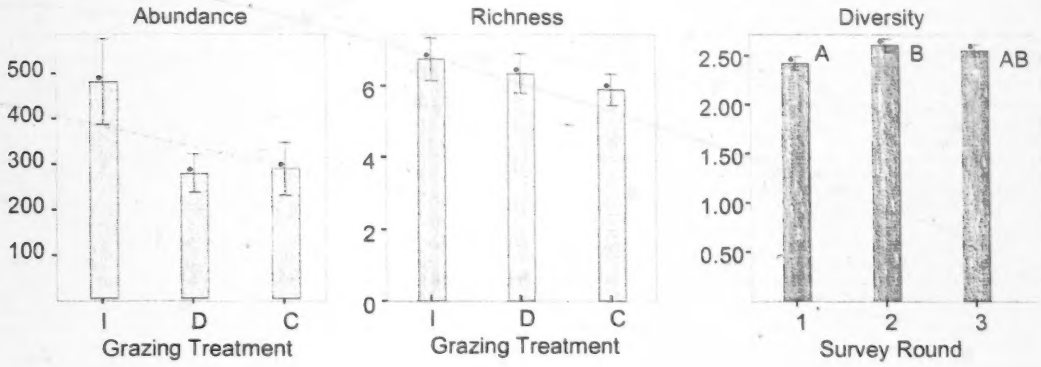
4.5.1 Grazing treatment and wetland aquatic macro-invertebrate community

No significant differences in macro-invertebrate abundance among grazing treatments were found overall for any specific aquatic taxa, when considering sampling round (no significant treatment effect, sampling period effect, or interaction; Figure 4-3). No significant differences were found in overall aquatic macro-invertebrate taxon richness or diversity among treatments when considering sampling round, however diversity was significantly higher in Round 2 compared to Round 1 (significant sampling period effect; no significant treatment or interaction; Table 4-1, Figure 4-3).

Table 4-1: Results for differences in aquatic macro-invertebrate diversity using Friedman's test.
(?^{2} values from Table B.1 in Zar 1999)

Source	df	H	?$^2_{crit}$	p-value
Treatment	2	5.051	5.991	not significant
Round	2	7.026	5.991	<0.05
Interaction	4	<1	9.488	not significant

Figure 4-3: Differences in wetland aquatic macro-invertebrate community among grazing treatments.
(Bars represent ±1 S.E.; alpha 0.05; Shannon's Diversity Index)



4.5.2 Comparison of odonates to overall aquatic macro-invertebrate community

Significant positive correlations were found between overall aquatic macro-invertebrate abundance and odonates when grouped by *Enallagma* spp., Zygoptera, and overall odonate larvae abundance (Table 4-2). Gastropod abundance was the only aquatic macro-invertebrate taxon that was significantly positively correlated to larval odonate abundance, specifically *Enallagma* spp. ($r=0.431$, $p<0.05$). It is possible that the significant relationship detected between Gastropoda and odonates may be due to Type I Error since (with an alpha of 0.05 and 11 taxa included in the tests) it is expected that at least one taxon will show significant results. However, the Subclass Gastropoda does re-occur as a significant variable in other analysis (see following sections). Larval odonate diversity was significantly positively correlated to aquatic macro-invertebrate diversity (Table 4-2). No significant correlation was found between odonate richness and overall macro-invertebrate richness.

Table 4-2: Significant correlations between wetland odonate and aquatic macro-invertebrate communities.
(Spearman's rank correlation coefficient; a=0.05; diversity =Shannon's Diversity Index)

		Odonata Diversity	Odonata Abundance	Zygoptera Abundance	*Enallagma* spp. Abundance
Overall Aquatic Macro-invertebrate	**Diversity**	+0.632			
	Abundance		+0.470	+0.482	+0.570

The relationship between larval odonates and their prey was analysed by comparing larval abundance, richness, and diversity to that of potential prey taxa. Potential prey taxa were identified *a priori* according to research on larval odonate diets and included the following eleven taxa: Gastropoda, Acari, Amphipoda, Coleoptera (larvae), Diptera, Ephemoptera, Corixidae, Gerridae, Trichoptera, Oligochaeta, and Hirudinea (Corbet 1999). A significant positive correlation was found for the diversity of larval odonates and their potential prey ($r=0.633$, $p<0.01$). No significant correlation was found between larval odonates and their prey with respect to abundance or taxa richness.

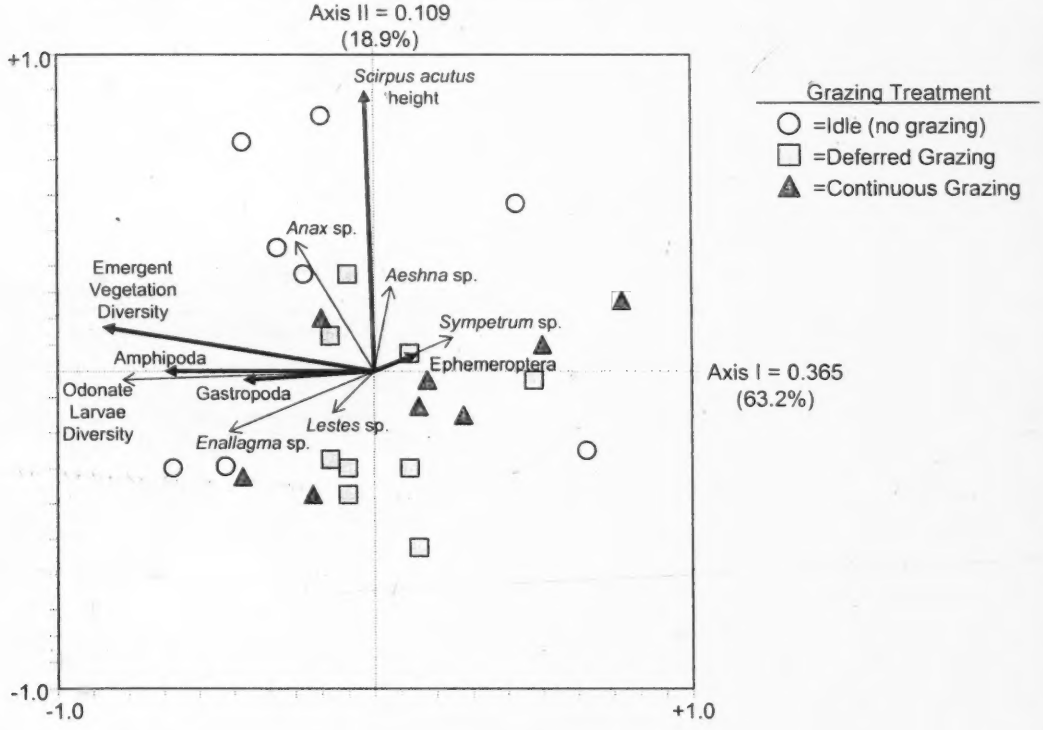
4.5.3 The odonate community in relation to the overall wetland environment

Larval and adult odonates were treated separately in direct ordination analysis since they represent two very distinct life stages. With respect to larval odonates (Figure 4-4), the first two ordination axes explained 82.1% of the total variation in both the species and the environmental parameters data set, and the first axis represented a strong environmental gradient (i.e. first eigenvalue is fairly high). Two

sites were confirmed as outliers in preliminary bi-plots, and deleted from the final ordinations because they were not representative of the target population (McCune & Grace 2002); Lomond Upslope East (continuous treatment) was directly influenced by an irrigation canal due to a breach in the embankment, and Backflood South (idle treatment) experienced a severe dewatering for the majority of the summer. Removal of these outliers did not notably change the results of the ordination (eigenvalues, results of the Monte-Carlo randomization, or relative tri-plot loci), but merely expanded the ordination space of interest.

The height of the emergent plant *Scirpus acutus* (great bulrush) represented a strong environmental gradient (i.e. large vector, closely associated with the second canonical axis) that mostly influenced the abundance of the larger Anisoptera larvae (i.e. *Aeshna* spp., *Anax* spp., and *Sympetrum* spp.). Smaller Zygoptera larvae abundance was more strongly related to emergent vegetation diversity, as well as Amphipoda and Gastropoda abundance. The abundances of Zygoptera and Anisoptera larvae were divergent. Larval odonate diversity was most strongly associated with emergent vegetation diversity, as well as Amphipoda and Gastropoda abundance. Grazing treatment was only weakly associated with larval odonate diversity, especially for deferred and continuously grazed wetlands. Grazing treatments were well scattered throughout the plot, but most grazed wetlands were not associated with *Scirpus acutus* height.

Figure 4-4: Relationship of emergent vegetation and aquatic macro-invertebrate abundance to the larval odonate community at prairie wetlands. (RDA, inter-species focus, bi-plot scaling; sum of eigenvalues = 0.577 or 97.7% of variation; 1st axis and sum of axes p=0.0001; two outliers (1 idle, 1 continuous) omitted from ordination)



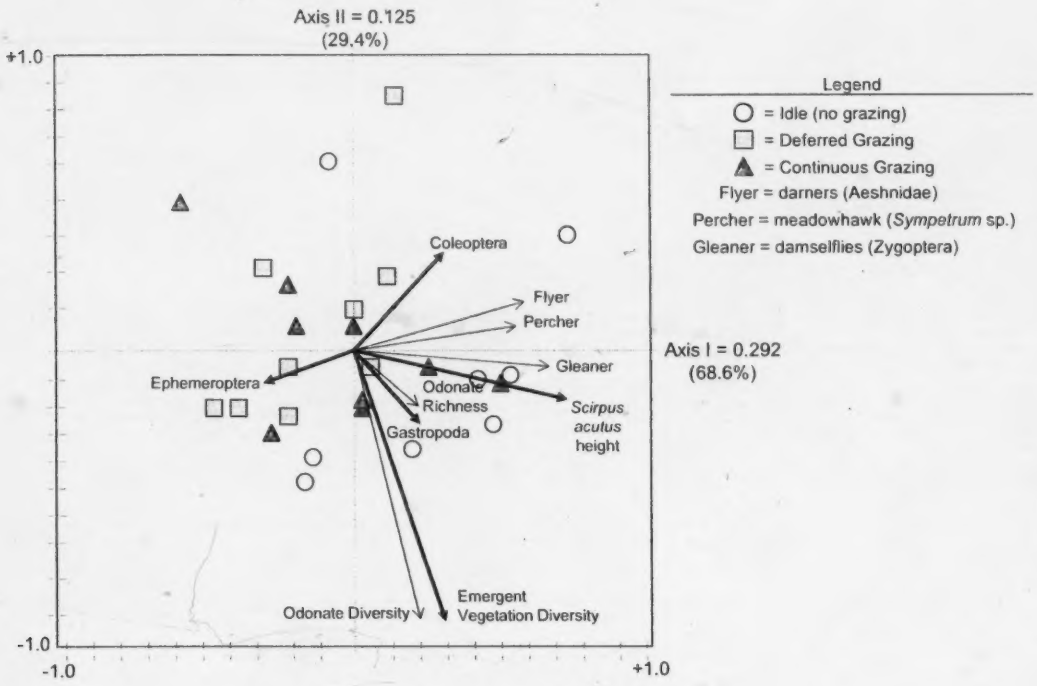
Two ordinations were performed on adult odonate data: 1) all twenty odonate species treated separately, and 2) species categorized by functional feeding groups. The twenty odonate species plot is cluttered; however it is important to show the variation within each feeding group as exhibited by individual species.

The first ordination axis (Figure 4-5) was a strong gradient and the ordination axes together explained 98.0% of the total variation in both the species and the environmental parameters data set for odonates characterized as Flyers, Perchers, and

Gleaners. One site was confirmed as an outlier in a preliminary ordination bi-plot and deleted from the final ordinations (McCune & Grace 2002); Backflood South (idle treatment) experienced severe de-watering for the majority of the summer. Flyer and Percher abundances were more closely related to each other than to Gleaner abundance, and the abundance of each feeding group contrasted with Ephemeroptera abundance. Odonate diversity was strongly associated with emergent vegetation diversity, and the abundance of all odonate feeding groups (especially Gleaners) was strongly associated in a positive direction with *Scirpus acutus* height.

Grazing treatments were only weakly associated with emergent vegetation diversity or odonate diversity. Most grazed wetlands were not at all or only weakly associated with odonate abundance or any aquatic macro-invertebrate abundance, with the exception of Order Ephemeroptera.

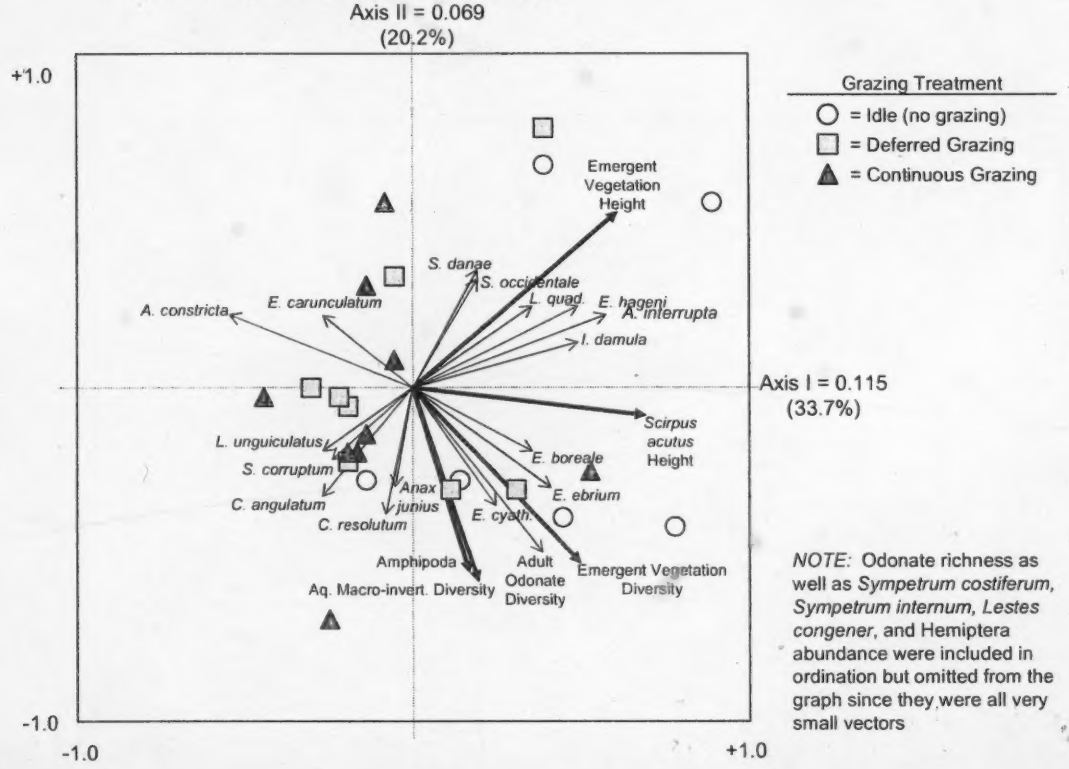
Figure 4-5: Relationship of emergent vegetation and aquatic macro-invertebrate abundance to adult odonate feeding groups at prairie wetlands. (RDA, inter-species focus, bi-plot scaling; sum of eigenvalues = 0.426 or 99.9% of variation; 1st axis p=0.0119, and sum of axes p=0.0016; one outlier (idle site) omitted from ordination)



When each of the twenty adult odonate species was treated separately, the two ordination axes only explained 53.9% of the variation in both the species and the environmental parameters data set and the strength of the gradient from the first ordination axis was diminished to 33.7% (Figure 4-6). Again, two outliers severely influenced the data and were deleted from the final ordinations; both Newell Main Dam and Backflood South (idle treatments) experienced severe draw-downs for a large portion of the summer. The abundance of every odonate species was somewhat associated with emergent vegetation height (especially the larger dragonflies) or

diversity (especially the smaller damselflies), except for *Aeshna constricta* (Lance-tipped Darner) and *Enallagma carunculatum* (Tule Bluet). The abundances of Zygopterans and Anisopterans diverged. The abundances of *Enallagma boreale, E. cyathigerum,* and *E. ebrium* were most strongly related to emergent vegetation diversity. Odonate diversity was strongly associated with emergent vegetation diversity, and to a lesser degree aquatic macro-invertebrate diversity and Amphipoda abundance. No specific grazing treatment was strongly associated with emergent vegetation diversity; however grazed wetlands (deferred or continuous) were not very strongly associated with taller *S. acutus*, or the overall height of emergent vegetation.

Figure 4-6: Relationship of emergent vegetation and aquatic macro-invertebrate abundance to twenty adult odonate species at prairie wetlands.
(RDA, inter-species focus, bi-plot scaling; sum of eigenvalues = 0.341 or 86.7% of variation; 1st axis p=0.0210, sum of axes p=0.0020; two outliers (idle sites) omitted from ordination)



4.6 Discussion

4.6.1 Aquatic macro-invertebrates and cattle grazing

The peak in aquatic macro-invertebrate diversity coincides with mid-summer (i.e. mid-July) and is consistent with peaks in diversity for wetland odonates (i.e. larval odonate diversity peaks in mid-July (Figure 2-9); adult odonate diversity peaks twice over the summer; once in early July and again in early-August (Figure 3-18).

Surrounding land use, in addition to hydroperiod and salinity, are reported to be important determinants of the aquatic invertebrate community in prairie wetlands (Zimmer et al. 2000); however grazing treatment showed no distinct impact on wetland macro-invertebrates (Figure 4-3). Grazing treatment did not affect the aquatic macro-invertebrate diversity, taxa richness, or abundance (i.e. overall or the abundance of any specific taxon).

The dissimilarity in the response of odonates and other aquatic macro-invertebrate taxa to wetland grazing regime is most likely due to the terrestrial adult life phase of odonates. Suitable odonate habitat must encompass requirements for all life stages (i.e. egg, larvae, and adult), and the terrestrial nature of adults makes odonates more closely connected to the emergent and terrestrial vegetation surrounding wetlands than other strictly aquatic macro-invertebrates (Corbet 1999). Since cattle are, at the very simplest, vegetation altering machines it makes sense that organisms, such as adult odonates, that are dependent on emergent vegetation structure would be sensitive to a reduction in vegetation by cattle herbivory, whereas organisms that are not dependent on emergent/terrestrial vegetation, such as other

more truly aquatic macro-invertebrates, would not be as sensitive to vegetation altering cattle.

4.6.2 Comparing wetland odonate community to other aquatic macro-invertebrates

A significant correlation exists for aquatic macro-invertebrates and Genus *Enallagma*, Suborder Zygoptera, and Order Odonata but the strength of this correlation decreases with each increase in odonate taxonomic level (i.e. less precise identification). Aquatic macro-invertebrate abundance is most strongly positively correlated with *Enallagma* spp. abundance ($r=^{+}0.570$, $p<0.05$). Aquatic vegetation is most likely the link between aquatic macro-invertebrate and *Enallagma* spp. larval abundance, since adults of the three most common *Enallagma* spp. (i.e. *E. boreale, E. cyathigerum,* and *E. ebrium*) are positively correlated to emergent vegetation diversity (Figure 4-6), as are most aquatic macro-invertebrate taxa (Figure 4-4 to 4-6).

The positive correlation between larval odonate diversity and aquatic macro-invertebrate diversity is quite strong ($r=^{+}0.632$, $p<0.01$). Aquatic vegetation is most likely the link to this association, and other studies show that increased structural heterogeneity in aquatic vegetation also is positively correlated with an increase in aquatic macro-invertebrate abundance (Zimmer et al. 2000, Painter 1999, Gilinsky 1984, Voigts 1976). Odonates and other aquatic macro-invertebrates share much of the same basic habitat requirements (i.e. 'clean' water, available prey, and substrate for concealing themselves from predators/prey) (Merritt and Cummins 1996). It follows that conditions that are favourable for increasing the number of odonate

species inhabiting a wetland (i.e. more emergent vegetation substrate) will also be benefit other aquatic macro-invertebrate taxa. As well, because odonates are a top aquatic predator, their habitat should be improved by the presence of other invertebrate prey.

The positive correlation between the diversity of larval odonates and potential prey items is almost identical to that of overall aquatic macro-invertebrates (i.e. $r=^{+}0.633$, $p<0.01$ for potential prey, and $r=^{+}0.632$, $p<0.01$ for overall macro-invertebrates). This supports the assumption that larval odonates at prairie wetlands are opportunistic feeders whose diets reflect ambient aquatic macro-invertebrate fauna (Corbet 1999). Larval odonate prey taxa were identified in this study based on results from other research rather than direct analysis of larvae gut contents or fecal pellets; therefore, the treatment of potential prey in this study is imprecise since I did not consider the influence of handling time or counter-adaptations by prey, and did not include Subclasses Ostrocoda or Cladocera in my invertebrate surveys, which (together with chironomids) are considered primary prey items for larval odonates (Corbet 1999). Size class (not measured in this study) may be important to consider since it may affect handling time as well as the tendency for the odonate to strike out in an attempt to catch and eat the invertebrate. Positive correlations among diversity of related taxa has been corroborated by other studies (Pearson & Cassola 1992, Kremen 1994, Flather et al. 1997, Pearson & Carroll 1998, Anderson & Vondracek 1999), and forms the basis of biodiversity indicator species theory. The strong correlation between larval odonate and aquatic macro-invertebrate communities suggests that the relative abundance and diversity of aquatic invertebrates, which are

more time/effort consuming to sort and identify, may instead be accurately inferred from the diversity of larval odonates.

4.6.3 The odonate community in relation to the overall wetland environment

When interpreting an ordination graph it is important to consider the distance and angle of the vectors. I chose to preserve the relationship, among species (i.e. focus on inter-species distances) rather than sites since I am most interested in the impact of environmental variables on the odonate community. With the focus on species, the species vectors in the ordination space are interpreted based on their relationship to the ordination axes, environmental vectors, and other species vectors (i.e. longer vectors and smaller angles between the species vector and axis or environmental/species vector indicate a stronger relationship), as well as to the sample sites (i.e. where the site falls at 90° to the species vector with longer distances indicating a stronger relationship).

Grazing treatment was not selected as a significant environmental parameter, and the association between grazing treatment and odonate abundance or diversity is weak (Figure 4-4 to 4-6). Grazed and ungrazed sites were roughly distributed as two clusters; idle sites were more closely positively associated with tall/diverse emergent vegetation, and abundant/diverse odonates and other aquatic macro-invertebrates than grazed sites (Figures 4-4 to 4-6). It may seem incongruous that grazing treatment was not significantly associated with odonate abundance or diversity, however *a priori* directed statistical analysis showed that grazing treatments significantly impact vegetation structure (Figure 3-6), vegetation structure is strongly positively associated

with odonate abundance (Figures 3-7 and 4-4 to 4-6), and both adult and larval odonate abundance are significantly lower at deferred grazed compared to ungrazed wetlands (Figure 2-6 and 3-15). The weaker association between grazing treatment and the odonate community most likely has more to do with my simplified treatment of cattle grazing as a categorical variable rather than a continuous variable.

The reliability of an ordination is assessed by considering the consistency of a given pattern, the proportion of variance represented (30-50% considered robust *a priori*), and the statistical strength of that pattern (*a priori* a=0.05) (McCune & Grace 2002). Interestingly, forward-selection highlighted emergent vegetation diversity and *S. acutus* height as the most important factors influencing separation along the first two axes for all ordinations (Figure 4-4 to 4-6). Emergent vegetation diversity and height were consistently roughly orthogonal on all three tri-plots, indicating that no matter how the odonate data are taxonomically delineated, vegetation structure and diversity remained the two most influential environmental parameters for the wetland odonate community. Odonate diversity in particular was always strongly associated with emergent vegetation diversity. Many of the patterns observed in the ordination tri-plots were found *a priori* in more directed and statistically robust analysis in previous chapters, further supporting my conclusions regarding these patterns (e.g. positive relationship between vegetation diversity and odonate diversity; positive relationship between vegetation height and odonate abundance; lack of significant water quality parameters).

The abundances of Anisoptera larvae and Zygoptera larvae were negatively related, especially with respect to *Sympetrum* spp. and *Enallagma* spp., which were

the two genera most commonly identified in the study area (Figure 4-4). Anisoptera larvae are large voracious predators that will eat smaller Zygoptera larvae (Corbet 1999); this predation pattern may explain an inverse relationship of abundance.

The structure and diversity of emergent vegetation is critically important to both adult and larval odonates (Figures 4-4 to 4-6). Emergent vegetation provides critical emergence structures for larval odonates and offers protection from predators and wind damage during the most vulnerable stage of their life cycle, as well as a wind-break for weaker flyers such as Zygoptera (Corbet 1999). Each genus of larval odonate identified in this study was characterized as 'climbers' that cling to submerged vegetation (Needham et al. 2000, Corbet 1999). The submerged portions of emergent plants (i.e. stems and roots) added to the structural diversity of larval odonate micro-habitat and provided a substrate on which larvae clung, to avoid detection from predators/prey. Additionally, tall lush emergent vegetation may serve to shade and regulate the water temperature in the littoral zone.

The significance of emergent vegetation height (specifically *S. acutus*) to Anisoptera larvae is most likely a reflection of suitable habitat for territorial or ovipositing adults (Figure 4-4). Adult Anisopterans make up the groups Flyers (i.e. large dragonflies that catch prey in flight; includes *Aeshna* spp., *Anax* spp., and *Libellula spp.*) and Perchers (i.e. medium sized dragonflies that dart out from perches; such as *Sympetrum* spp.). Odonates from both of these groups use the tips of tall emergent vegetation, such as *S. acutus*, as perches from which they defend territories, acquire mates, or catch prey (Corbet 1999, Needham et al. 2000). Ovipositing

Aeshna spp. and *Anax* spp. will saw into the stems of emergent plants and deposit their eggs inside plants (Corbet 1999).

Larval Zygoptera abundance was associated with emergent vegetation diversity rather than *S. acutus* height (Figure 4-4). This association most likely also reflects habitat requirements of adult Zygopterans since these weak flyers tend to select sites low amongst the emergent vegetation while searching for prey and oviposition sites (Corbet 1999). Other studies have found positive correlations between the abundance and richness of aquatic vegetation and larval odonates (Muller et al. 2002, Painter 1999, Sahlen and Ekestubbe 2001). More diverse emergent vegetation provided a more heterogeneous habitat for adult Zygopterans, and was reflected in abundance patterns for *Enallagma* spp., as well as *Ischnura* spp. and *Coenagrion* spp. (Figure 4-6). Adult *Enallagma* spp. are dependent on wetland vegetation since all species are either epiphytic or endophytic ovipositors (i.e. deposit eggs either on or within vegetation; Corbet 1999), whereas adult *Ischnura* sp are well-known for hiding among emergent vegetation stems (Westfall and May 1996). The positive correlation between Zygoptera larvae abundance and Gastropoda/Amphipoda abundance is likely explained by a shared dependency on diverse aquatic vegetation, which is a source of food for Gastropoda and Amphipoda (Merritt and Cummins 1996; Figure 4-4).

The aquatic macro-invertebrate community was positively related to emergent vegetation diversity, especially for Subclass Gastropoda and Order Amphipoda (Figures 4-3 to 4-5). Diverse aquatic vegetation positively impacts aquatic invertebrate abundance in this and other studies (Zimmer et al. 2000) since aquatic

vegetation provides cover from visual predators (Crowder & Cooper 1982) and increases food resources for aquatic invertebrates (Carpenter & Lodge 1986).

Gastropoda and Ephemeroptera recur as taxa that significantly contribute to patterns in the wetland odonate community (Figure 4-4 to 4-6), and both taxa are suggested in the literature as suitable biological indicators (US EPA 2002b, Merritt and Cummins 1996). The abundance of Gastropoda, implicated as an indicator of lentic habitat (US EPA 2002b), is positively correlated with Zygopteran abundance, as well as odonate diversity and richness. Conversely, Ephemoptera abundance is consistently negatively correlated with odonate abundance and diversity, as well as all other environmental parameters. This suggests that the suitability of Order Ephemeroptera as an indicator of habitat quality in lotic ecosystems does not extend to lentic ecosystems. Differences may be due to the inherent differences between lotic and lentic ecosystems, since lotic systems are typically cooler nutrient poor waters with higher oxygen levels while lentic systems (especially prairie wetlands) are typically warmer nutrient-rich environments (Murkin et al. 2000). The vast majority of Ephemeroptera identified in this study were either *Caenis* spp. (50%) or *Sipholonurus* spp. (49%) that flourish in the silty sediments of hyper-eutrophic lentic habitats; these are distinctly different from sensitive clear-water specialists used in rapid assessments of lotic habitats (Merrit & Cummms 1996).

4.7 Summary and recommendations

Larval odonate diversity was positively correlated with larval prey and overall aquatic macro-invertebrate diversity, and odonate larval abundance was positively

correlated with overall aquatic macro-invertebrate abundance, which supports the hypothesis that larval odonate diversity is an accurate biodiversity indicator of the aquatic macro-invertebrate diversity at prairie wetlands (i.e. accept more specific Hypothesis #5).

Cattle grazing significantly decreased wetland vegetation structure (i.e. height), but had no biologically significant effect on wetland water quality or vegetation abundance (i.e. % cover), richness, or diversity. My results support the hypothesis that odonates (larval and adult) represent accurate biological indicators of the impact of cattle grazing on vegetation structure and aquatic macro-invertebrate community at prairie wetlands (i.e. accept more specific Hypothesis #6). Surveying the larval and adult odonate community of a wetland provides information that is inherently important, as well as ecologically insightful as to the overall quality of prairie wetland habitat for odonates and other harder-to-survey aquatic invertebrates. Furthermore, aquatic macroinvertebrates are an essential part of wetland trophic webs and serve as a conduit of energy flow from aquatic systems to the terrestrial systems via birds and other vertebrates (Cox et. al. 1998).

Cattle grazing was treated very simplistically in this study; grazing treatments were defined based on the duration of cattle at a site (i.e. all summer vs. part of summer vs. control treatment), without directly addressing the absolute magnitude of this effect on flora (i.e. consider pasture size, standing crop, productivity, density of wetlands in pasture, frequency of grazing, and herd size). Odonates responded to changes in wetland vegetation, but few direct relationships between odonate community and grazing treatment, as defined by idle, continuous, or deferred, were

found. Impacts of cattle grazing are typically site specific, and future studies that address cattle grazing as a more complex concept may provide a clearer picture of its ecological impacts on the odonate community. This study has demonstrated positive correlations between odonates and wetland vegetation structure and aquatic macro-invertebrate abundance and diversity, and highlighted odonates as a suitable habitat, biodiversity, and population indicator species, allowing future studies to focus on developing more specific and quantitative Indexes of Biotic Integrity (IBI's) for wetland systems, similar to those widely used in lotic systems. Future studies should consider the use of multivariate analysis rather then univariate analyses to better clarify and identify variables that significantly affect the odonate community of a wetland.

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Chapter 5

Synthesis of Chapters: Are Odonates Suitable Biological Indicators?

5.1 Summary

Biological indicators are scientifically valid when selections are based on scientific criteria and when empirical studies confirm correlations between the indicator and the indicatee (McGeoch 1998). Odonates are well suited as indicator taxa (Table 1-1) and this study established correlations between the odonate community, and the vegetation and aquatic invertebrate communities of prairie wetlands (Table 5-1).

Wetland vegetation was the most important environmental parameter influencing odonate abundance, reproductive effort, and diversity, as well as an important ecological link explaining correlations between odonate and other aquatic macro-invertebrate communities. The direct impact of cattle grazing on wetland vegetation and the strong correlation between vegetation structure/diversity and odonate abundance/diversity begs the question: why not simply measure wetland vegetation to infer the condition of the odonate community? Surveying the wetland vegetation community gives information regarding its present condition; surveying long-lived riparian obligate taxa, such as odonates, provides additional temporal information such as the impact of past events no longer detectable in the vegetation community (i.e. intermittent events like short-term intense grazing). Maximum ecological insight is gained when bioindicators are monitored in concert with relevant environmental data (Faith & Walker 1996).

Table 5-1: Suitability of odonates as biological indicators at prairie wetlands.

Hypothesis #	Criteria for a suitable bioindicator	Significant?	Conclusion	Detailed Findings
2 (page 17)	Sensitive to changes to wetland water quality due to cattle grazing	No	Inconclusive	Nutrient inputs from cattle did not significantly impact wetlands water quality.
4 (page 17)	Sensitive to changes to wetland habitat due to cattle grazing	Yes	Suitable habitat indicator	Cattle grazing significantly decreased the height of emergent vegetation. Odonates were sensitive to this change with decreased abundance, diversity, and (Zygoptera) reproductive effort at wetlands with shorter emergent vegetation.
5 (page 18)	Sensitive to changes in aquatic macro-invertebrate community of a wetland	Yes	Suitable population indicator	Odonate abundance is positively correlated to overall aquatic macro-invertebrate abundance
5 (page 18)	Sensitive to changes in aquatic macro-invertebrate diversity	Yes	Suitable biodiversity indicator	Odonate diversity is positively correlated to aquatic macro-invertebrate diversity
6 (page 18)	Consistently and sensitive to cattle impacts at prairie wetlands when environmental and species data are analysed systematically	Yes	Suitable habitat and biodiversity indicator	Emergent vegetation height and diversity consistently and strongly influence larval and adult odonate abundance and diversity. Aquatic macro-invertebrate diversity is positively correlated to emergent vegetation and odonate diversity. The abundance of key aquatic macro-invertebrate taxa are positively correlated to odonate abundance and diversity.

Grazing treatments significantly impacted wetland vegetation, which in turn, significantly impacted larval and adult odonates, but grazing treatments did not often directly impact wetland odonates. This apparent disconnect is most likely due to the crude measure of cattle grazing as idle, deferred, or continuous rather than

a more precise quantitative metric. Treating grazing regime as a categorical value combined with the logistical constraints of being unable to alter established grazing regimes meant that factors critical to determining the utilization of a pasture/wetland by cattle were not taken into account (i.e. number of cattle per pasture, size of pasture, number of wetlands per pasture, number of days cattle present).

This study is well suited for testing the Intermediate Disturbance Hypothesis (IDH) which contends that diversity is lowest at sites with either extreme (i.e. continuously grazed wetlands) or minor (i.e. ungrazed wetlands) disturbance (Connell 1978). Differences in odonate diversity among treatments were contradictory for adult and larval odonates; adult diversity was not significantly impacted by grazing treatment (Figure 3-18) while the patterns of larval diversity among treatments supported the IDH (Figure 2-9). Cattle significantly impacted wetland vegetation structure (Chapter 3) but had little impact on wetland water quality (Chapter 2). Results suggest that cattle impact larval odonates via the removal of predatory refugia (i.e. emergent vegetation) or by altering ovipositing habitat, and that although adult odonates are present at wetlands, they are not necessarily encountering ovipositing habitat and depositing eggs at that site. Therefore, this study supports the IDH when considering the breeding portion of a wetland odonate community (i.e. larval odonates), since diversity is consistently lower at chronically disturbed sites, and highest at wetlands immediately following a short-term acute disturbance.

I can not justify a list of specific odonate species that must be present or absent to assess the quality of prairie wetland habitat; however, this study confirms that odonates are well suited for indicating the impact of grazing disturbance at these

dynamic systems. The strongest relationships with wetland biota were often manifested in Suborder Zygoptera (i.e. Gleaners) yet I recommend that all odonate taxa be included in future wetland monitoring programs since the use of multi-species assemblages as bioindicator taxa guards against over-simplifying complex systems and allows for a broader scale of inference, which is more constructive in the field of conservation biology (Wilson 1988, Croonquist & Brooks 1991, Kovacs et al. 1992, Caro & O'Doherty 1999).

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Appendix–A: Literature search for the history of biological indicator theory

Results from search of JSTOR© database search for "biological indicator(s)" OR "bio indicator(s)" OR "bioindicator(s)" OR "surrogate species" OR "indicator species" OR indicator taxa" were added to a database and categorized according to year of publication, biological indicator taxa (i.e. aquatic macro- and micro-invertebrates, fish and amphibians, birds and mammals, terrestrial invertebrates, diatoms, protozoa, and plants), and the environmental condition being assessed (i.e. terrestrial or aquatic habitat quality, soil chemistry, fire history, or climate change). Graphs were produced using SPSS 11.0 for Windows. The scientific journals included in the search are listed alphabetically below:

- Abstracts of the Papers Communicated to the Royal Society of London
- Abstracts of the Papers Printed in the Philosophical Transactions of the Royal Society of London
- American Journal of Botany
- American Midland Naturalist
- American Naturalist
- Annals of the Missouri Botanical Garden
- Annual Review of Ecology and Systematics,
- Biodiversity Letters
- Biotropica
- Botanical Bulletin
- Botanical Gazette
- Brittonia
- Bulletin of the Torrey Botanical Club
- Conservation Biology
- Diversity and Distributions
- Ecological ApplicationsInternational Journal of Plant Sciences,
- Ecological Monographs,
- Ecology
- Evolution
- Functional Ecology,
- Global Ecology and Biogeography Letters
- Isis
- Journal of Animal Ecology
- Journal of Applied Ecology
- Journal of Biogeography
- Journal of Ecology
- Journal of the Torrey Botanical Society
- Journal of Tropical Ecology
- Limnology and Oceanography
- Midland Naturalist
- Missouri Botanical Garden Annual Report

- New Phytologist
- Paleobiology
- Philosophical Transactions (1665-1678)
- Philosophical Transactions (1683-1775)
- Philosophical Transactions of the Royal Society of London
- Philosophical Transactions of the Royal Society of London. A
- Philosophical Transactions of the Royal Society of London. B
- Philosophical Transactions of the Royal Society of London. Series A. Mathematical and Physical Sciences
- Philosophical Transactions of the Royal Society of London. Series B. Biological Sciences
- Philosophical Transactions of the Royal Society of London. Series A. Containing Papers of a Mathematical or Physical Character
- Philosophical Transactions of the Royal Society of London. Series B. Containing Papers of a Biological Character
- Philosophical Transactions: Biological Sciences
- Philosophical Transactions: Mathematical
- Philosophical Transactions: Physical Sciences and Engineering
- Philosophy of Science
- Physical and Engineering Sciences
- Proceedings of the National Academy of Sciences of the United States of America
- Proceedings of the Royal Society of London
- Proceedings of the Royal Society of London. Series A. Containing Papers of a Mathematical and Physical Character
- Proceedings of the Royal Society of London. Series A. Mathematical and Physical Sciences
- Proceedings of the Royal Society of London. Series B. Biological Sciences
- Proceedings of the Royal Society of London. Series B. Containing Papers of a Biological Character
- Proceedings: Biological Sciences
- Proceedings: Mathematical and Physical Sciences,
- Proceedings: Mathematical, Physical, and Engineering Sciences
- PSA: Proceedings of the Biennial Meeting of the Philosophy of Science Association
- Quarterly Review of Biology
- Science
- Science Studies
- Scientific Monthly
- Social Studies of Science
- Systematic Biology
- Systematic Botany
- Systematic Zoology

Appendix–B: List of vegetation detected at study sites in August 2000.

	Genus species	Common Name	Native plant?
1	*Achillea millefolium*	common yarrow	Y
2	*Agropyron smithii*	western wheat grass	N
3	*Alisma gramineum*	narrow-leaved water plantain	Y
4	*Antennaria sp.*	pussy-toes (or everlasting)	Y
5	*Artemesia sp.*	sagebrush sp.	Y
6	*Artemesia cana*	silver sagebrush	Y
7	*Artemesia frigidara*	pasture sagewort	N
8	*Aster ericoides*	tufted white prairie aster	Y
9	*Aster falcatus*	creeping white prairie aster	Y
10	*Beckmannia syzigachne*	slough grass	Y
11	*Caliciales (Order)*	lichen sp.	?
12	*Carex sp.*	sedge sp.	Y
13	*Ceratophyllum sp.*	coontail (common hornwort)	Y
14	*Cirsium arvense*	Canada thistle	N
15	*Eleocharis palustris*	common spikerush	Y
16	*Eleocharis sp.*	spikerush sp.	Y
17	*Elodea canadensis*	Canada waterweed	Y
18	*Hordeum jubatum*	foxtail	N
19	*Juncus balticus*	wire rush	Y
20	*Lemna minor*	common duckweed	Y
21	*Lemna trisulca*	ivy duckweed	Y
22	*Lycopodium sp.*	prairie club-moss sp.	Y
23	*Mentha arvensis*	wild mint	Y
24	*Monolepis nuttalliana*	goosefoot	N
25	*Myriophyllum sp.*	Milfoil sp.	Y
26	*Phalaris arundinacea*	reed canary grass	Y
27	*Plantago major*	common plantain	N
28	*Poa sp.*	bluegrass sp.	N
29	*Polygonum amphibium*	water smartweed	Y
30	*Potamogeton pectinatus*	sago water weed	Y
31	*Potamogeton richardsoni*	Richardson's pondweed	Y
32	*Potamogeton sp.*	pondweed	Y
33	*Potentilla anserina*	silverweed	Y
34	*Rannunculus sp.*	buttercup sp.	Y
35	*Ratibida columnifera*	prairie cone-flower	Y
36	*Riccia fluitans*	riccia	N
37	*Rosa woodsii*	common wild rose	Y
38	*Rumex occidentales*	western dock	Y
39	*Sagittaria sp.*	Arrowhead sp.	Y
40	*Salicornia rubra*	red glasswort	Y
41	*Salix sp.*	willow sp.	Y
42	*Scirpus acutus*	great bulrush	Y
43	*Scolochloa festucacea*	link (whitetop)	Y
44	*Solidago canadensis*	Canada goldenrod	Y
45	*Sonchus sp.*	sow thistle	N
46	*Sparganium sp.*	burr reed	Y
47	*Symphoricarpos sp.*	snowberry	Y
48	*Taraxacum sp.*	dandelion sp.	N
49	*Tragopogon dubius*	goat's-beard	N
50	*Trifolium sp.*	clover sp.	N
51	*Typha latifolia*	cattail	Y
52	unknown	filamentous algae	?
53	unknown	fungus sp.	?
54	unknown	planktonic algae	?
55	*Vicia cracca.*	tufted vetch	N

Appendix–C: Taxonomic level of identification for aquatic macro-invertebrates collected at study sites during 2001 (using Clifford 1991).

Phylum	Class	Subclass	Order	Family	Genus	species
Annelida	Clitellata	Hirudinea	Pharyngobdellida	Erpobdellidae	*Dina or Mooreobdella*	-
Annelida	Clitellata	Hirudinea	Pharyngobdellida	Erpobdellidae	*Erpobdella*	-
Annelida	Clitellata	Hirudinea	Pharyngobdellida	Erpobdellidae	*Nephelopsis*	-
Annelida	Clitellata	Hirudinea	Rhynchobdellida	Glossiphonidae	*Glossiphonia*	-
Annelida	Clitellata	Hirudinea	Rhynchobdellida	Glossiphonidae	*Helobdella*	-
Annelida	Clitellata	Hirudinea	Rhynchobdellida	Glossiphonidae	*Theromyzon*	-
Annelida	Clitellata	Hirudinea	Gnathobdellidae	Hirudinidae	*Mollibdella*	-
Annelida	Clitellata	Oligochaete	-	Naididae	-	-
Annelida	Clitellata	Oligochaete	-	Lumbriculidae	-	-
Arthropoda	Arachnida	-	Acari	-	-	-
Arthropoda	Insecta	-	Amphipoda	-	*Gammarus*	*lacustrus*
Arthropoda	Insecta	-	Amphipoda	-	*Hyallela*	*azteca*
Arthropoda	Insecta	-	Coleoptera	Dryopidae	*Helichus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Agabus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Colymbetes*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Coptotomus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Dytiscus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Graphoderus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Hydroporus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Hygrotus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Hygrotus or Hydroporus*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Ilybius*	-
Arthropoda	Insecta	-	Coleoptera	Dytiscidae	*Rhantus*	-
Arthropoda	Insecta	-	Coleoptera	Haliplidae	*Haliplus*	-
Arthropoda	Insecta	-	Coleoptera	Haliplidae	*Peltodytes*	-
Arthropoda	Insecta	-	Coleoptera	Hydrophilidae	*Berosus*	-
Arthropoda	Insecta	-	Coleoptera	Hydrophilidae	*Hydrobius*	-
Arthropoda	Insecta	-	Coleoptera	Hydrophilidae	*Hydrophilus*	-
Arthropoda	Insecta	-	Coleoptera	Hydrophilidae	*Tropisternus*	-
Arthropoda	Insecta	-	Coleoptera	Hydrophilidae	*Laccobius*	-
Arthropoda	Insecta	-	Coleoptera	Gyrinidae	*Gyrinus*	-
Arthropoda	Insecta	-	Diptera	Anthomyiidae	-	-
Arthropoda	Insecta	-	Diptera	Ceratopogonidae	-	-
Arthropoda	Insecta	-	Diptera	Chaoboridae	-	-
Arthropoda	Insecta	-	Diptera	Chironomidae	-	-
Arthropoda	Insecta	-	Diptera	Culicidae	-	-
Arthropoda	Insecta	-	Diptera	Ephydridae	-	-
Arthropoda	Insecta	-	Diptera	Sciomyzidae	-	-
Arthropoda	Insecta	-	Diptera	Stratiomyidae	-	-
Arthropoda	Insecta	-	Diptera	Syrphidae	-	-
Arthropoda	Insecta	-	Diptera	Tanyderidae	-	-
Arthropoda	Insecta	-	Diptera	Tipulidae	-	-
Arthropoda	Insecta	-	Ephemeroptera	Ametropodidae	*Ametropus*	-
Arthropoda	Insecta	-	Ephemeroptera	Baetidae	*Baetidae*	-

Arthropoda	Insecta	-	Ephemeroptera	Caenidae	*Caenis*	-
Arthropoda	Insecta	-	Ephemeroptera	Siphlonuridae	*Analetris*	-
Arthropoda	Insecta	-	Ephemeroptera	Siphlonuridae	*Siphlonurus*	-
Arthropoda	Insecta	-	Hemiptera	Belostomatidae	-	-
Arthropoda	Insecta	-	Hemiptera	Corixidae	-	-
Arthropoda	Insecta	-	Hemiptera	Gerridae	-	-
Arthropoda	Insecta	-	Hemiptera	Mesoveliidae	-	-
Arthropoda	Insecta	-	Hemiptera	Notonectidae	-	-
Arthropoda	Insecta	-	Hemiptera	Saldidae	-	-
Arthropoda	Insecta	-	Hemiptera	Velidae	-	-
Arthropoda	Insecta	-	Lepidoptera	-	-	-
Arthropoda	Insecta	-	Trichoptera	Leptoceridae	*Mystacides*	-
Arthropoda	Insecta	-	Trichoptera	Leptoceridae	*Nectopshyche*	-
Arthropoda	Insecta	-	Trichoptera	Leptoceridae	*Oecetes*	-
Arthropoda	Insecta	-	Trichoptera	Leptoceridae	*Triaenodes*	-
Arthropoda	Insecta	-	Trichoptera	Limnephilidae	*Asynarchus*	-
Arthropoda	Insecta	-	Trichoptera	Limnephilidae	*Limnephilus*	-
Arthropoda	Insecta	-	Trichoptera	Phraganeidae	*Agrypnia*	-
Mollusca	Gastropoda		-	Lymnaeidae	*Lymnaea*	-
Mollusca	Gastropoda		-	Lymnaeidae	*Stagnicola*	-
Mollusca	Gastropoda		-	Physidae	*Physa*	-
Mollusca	Gastropoda		-	Planorbidae	*Helisoma*	-
Mollusca	Gastropoda		-	Planorbidae	*Promenetus*	-
Mollusca	Gastropoda		-	Valvatidae	*Valvata*	-
Mollusca	Pelecypoda		-	Sphaeriidae	*Psidium*	-
Mollusca	Pelecypoda		-	Sphaeriidae	*Sphaerium*	-